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P. O. Box 14222, Baghdad, Iraq.
College of Medicine
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E-mail: Iraqi_jms_alnahrain@yahoo.com

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3. Chapter in book: Phillips SJ, and Whisnant JP. Hypertension and strock. In: Laragh JH, and Brenner BM. editors. Hypertension: Pathophysiology, diagnosis, and management. 2nd ed. NewYork: Raven Press; 1995. p. 465-78.

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Editorial:

Newborn screening for inborn errors of metabolism (IEM)

Hala S. Arif *CABP*

Inborn errors of metabolism (IEM) are monogenic diseases resulting in deficient activity in a single enzyme in a pathway of intermediary metabolism. Sir Garrod, was the first to recognize heritable blocks in normal human metabolic flow that conformed to Mandelian mechanisms of inheritance, and gave the name **IEM**, at **1908**.

Single gene defects result in abnormalities in the synthesis or catabolism of proteins, carbohydrates, fats, or complex molecules. Most are due to a defect in an enzyme or transport protein, which results in a block in a metabolic pathway. Effects are due to toxic accumulations of substrates before the block, intermediates from alternative metabolic pathways, defects in energy production and use caused by a deficiency of products beyond the block, or a combination of these metabolic deviations. Nearly every metabolic disease has several forms that vary in age of onset (from few days after birth till adulthood), clinical severity, and, often, mode of inheritance.

The incidence collectively, is estimated to be approximately 1 in 4000 live births.

The international frequencies for each individual inborn error of metabolism vary. Of term infants who develop symptoms of sepsis without known risk factors, as many as 20% may have an inborn error of metabolism.

Dept. Pediatrics, College of Medicine, Al-Nahrain University.

There are over **300** gene disorders leading to specific biochemical defects of IEM,
a specific intervention that may prevent morbidity and/or is available in at least **1/3**$^{\mathbf{rd}}$ of the cases.

IEM may be implemented for sudden deterioration in a previously normal baby, of unexplained neonatal death. Older children with unexplained encephalopathy, mental retardation, epilepsy. Unexplained hepatomegaly, fulminant or chronic liver disease ending in cirrhosis, or at times hepatic carcinoma. Renal stones. Cardiomyopathy, rhabdomyolysis, maternal acute fatty liver of pregnancy.

Most of the diseases that exhibit clinical consequences manifest (or can be detected) in the newborn period or shortly after, on the other hand those that present early in the neonatal period are often lethal if proper treatment is not implemented, so early detection at least for some of these disorders has proven very effective in

treatment or management, avoiding later morbidity &/or mortality.

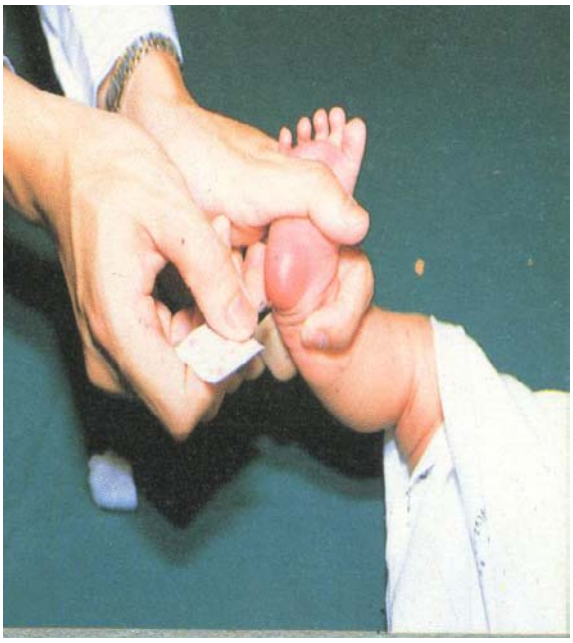
Newborn screening is the process of testing newborn babies for treatable metabolic,genetic, endocrinologic, and hematologic diseases. Robert Guthrie is given much of the credit for pioneering the earliest screening for phenylketonurea (one of the aminoacid metabolic disorders) in the 1960s using blood samples on filter paper obtained by pricking a newborn baby's heel on the second day of life to get a few drops of blood. Congenital hypothyroidism was the second disease widely added in the 1970s. The development of spectrometry screening by Edwin Naylor and others in the early 1990s led to a large expansion of potentially detectable congenital metabolic diseases reaching up to 30 diseases detected from a single blood spot sample, including most fatty acids, organic acids, aminoacids & urea cycle disorders, with a high sample throughput permitting the analysis of >100 samples in few hours. At present ,tandem mass spectrometry is being used as the screening technique for the diagnosis of IEM in newborns & sick infants in many clinical biochemical laboratories in USA, Europe, Australia & Japan, though the lists of screened diseases vary widely, according in part to the magnitude of the disease problem in that society.

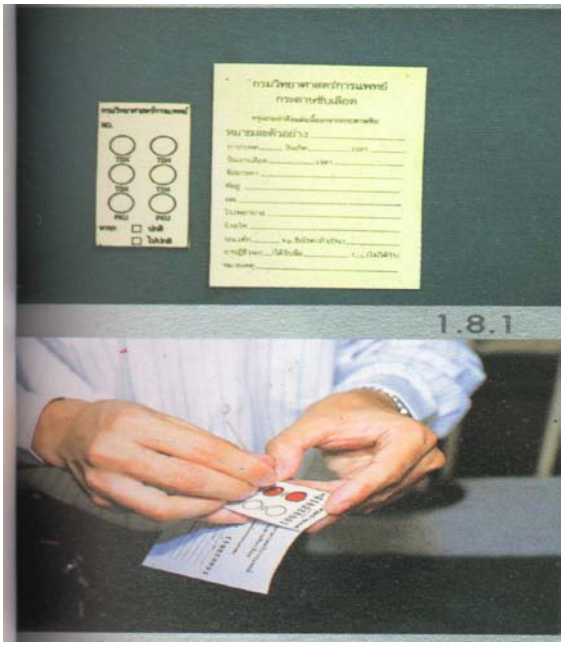
The current experiences in the Middle East and North Africa region (MENA), where the population is about 400 million, with high birth rate and an estimated 10 million newborns per year, the population is characterized by a high consanguinity (25-70%) and a high percentage of first-cousin marriages, so inherited metabolic disorders, neurogenetic disorders, Haemoglobin disorders and birth defects are relatively more common among this population, and numerous studies highlighted the need for newborn screening programs, despite that there is a slow progress in developing and implementing preventive genetic programs. There are only 4 countries that are executing national newborn screening. One of the earliest centers that applied tandem mass spectrometry in neonatal screening was the King Faisal Specialist Hospital & Research center in Riyadh / KSA that applied this technology since mid nineties.

In Iran, this technique started to be applied since 2002, in the Metabolic disease center in Tehran university, other modes of screening programmes were used since 1995, a second center was developed in Zanjan (Zanjan Metabolic Disease Center ZMDRC).

Lately, in Lebanon, international cooperation allowed the acquisition of this technology at the Newborn Screening Laboratory (NSL) of the Saint Joseph University (USJ) in the capital city of Beirut since 2006. NSL is currently screening up to 20% of all newborns in Lebanon.

In Iraq our Biochemical laboratories are not yet providing even the primary level of diagnostic tests with significant reliability, definitely they are not equipped yet with these newer diagnostic technologies , big steps need to be taken for developing national strategies for prevention and should learn from experiences at regional and international screening programs.

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Mineral Homeostasis in Preeclampsia

Faisal Gh. Al-Rubaye[1] *MBChB; MSc; PhD*, **Maha M. Al-Bayati**[2] *MBChB;CABOG*, **Tariq Hovthy Al-Khayat** *PhD*.

Abstract

Background: Preeclampsia is a form of high blood pressure manifested during pregnancy, it is a common major complication causing significant morbidity and mortality; however, its etiology is unknown. Moreover, data on mineral homeostasis and on cation pattern during pregnancy are conflicting. Also, the status of ionized calcium and magnesium during pregnancy and its complication preeclampsia have not been described adequately.

Objective: to demonstrate the pattern of minerals during preeclampsia with respect to normal pregnancy.

Subject and methods: the present study is a cross-sectional case-control study includes measurement of minerals (calcium and magnesium) in 60 patients with preeclampsia. They are classified into two groups according to gestational age:

- Preeclamptics in the second trimester G1: (n=30).
- Preeclamptics in the third trimester G2: (n=30,).

The results are compared with 60 apparently healthy pregnants controls. They are classified according to gestational age into two groups:

- Pregnants in the second trimester G3: (n=30).
- Pregnants in the third trimester G4: (n=30).

Results: show that serum corrected calcium and serum magnesium are significantly reduced in preeclamptics when compared with normal pregnants. In addition, there was a reduction in free calcium and free magnesium that was accompanied by a significant high elevation of the ratio between ionized calcium to ionized magnesium.

Conclusion: preeclamptics (in different gestational age groups) have altered mineral status when compared with healthy pregnants matched with their age and gestational age.

Key words: preeclampsia, calcium, magnesium.

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Introduction

Preeclampsia is defined as the onset of hypertension and the presence of proteinuria during pregnancy, usually occurring after the 20th week of gestation in a previously normotensive woman and resolving completely by the sixth week after delivery of fetus [1, 2].

The pathophysiology of preeclampsia is thought to represent a defective response to the physiologic demands of normal pregnancy [2, 3]. Normal pregnancy is associated with profound changes in maternal homeostasis [4]. The endpoint of these changes is to provide the fetus with the necessary environment for growth and the mother with adequate protection against pregnancy complications [4].

During normal pregnancy, maternal plasma total calcium concentrations fall, primarily because of the decrease in serum albumin to which the mineral is predominantly bound in the circulation and it seems likely that there is a relatively little change in unbound ionized calcium. However, there is a

[1]**Dept. Chemistry & Biochemistry,**[2]**Dept. Obstetrics & Gynecology, College of Medicine, Al-Nahrain University,**[3]**Dept. Biochemistry, College of Medicine, Babylon University**

Adress Correspondence to: Dr. Faisal Gh. Al-Rubaye.

E- mail: faisal3ghazi@yahoo.com

Mobile: 07702640792

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substantial fetal need for calcium [5]. It is now clear that the dynamics of calcium homoeostasis are in fact substantially altered in pregnancy [5]. pregnancy-induced hypomagnesemia has been reported previously. however; the status of ionized magnesium during pregnancy and its relation to other important cations such as ionized calcium have not been described adequately it is the "free" or ionized magnesium that exerts biological activity[6].

It was suggested that a deficiency in magnesium contributed to the development of vasoconstriction in preeclampsia [7]. Also, deficiencies in calcium intake have been linked to preeclampsia/eclampsia, and hypocalciuria and deviations in both 1,25-$(OH)_2D_3$ and PTH have been shown in women with preeclampsia[7].

Subjects & Methods

A-Subjects

The study was a cross-sectional, case-control study conducted on 60 patients with preeclampsia (PE) attending the Obstetric Consultant-Clinic, Antenatal Clinic, and Labor Ward at Al-Kadhimiya Teaching Hospital, for re-evaluation of newly diagnosed PE, or for delivery.

The diagnosis of PE was based on clinical criteria that were hypertension (absolute BP of 140/90 mmHg twice over 4 hr without prior comparison) [1, 2] and proteinuria (21.5 mg of urinary protein per mmol creatinine) [8].

The exclusion criteria, which were used for cases and controls, were gestational or chronic hypertension, diabetes mellitus, renal disease, multifetal gestation, intrauterine fetal death, and pregnancy less than 20 weeks of gestation.

Depending on the gestational age, the patients were divided into two groups:

1.Preeclamptics in the second trimester (G1):

Includes thirty Preeclamptics in their second trimester of pregnancy. Age range was from 18 to 37 years (mean age $\pm$ SD = 26.1 $\pm$ 6.4 year). The gestational age range was from 20 to 28 weeks (mean gestational age $\pm$ SD = 26.3 $\pm$ 1.5 week).

2.Preeclamptics in the third trimester (G2):

Includes thirty preeclamptics in their third trimester of pregnancy. Age range was from 18 to 40 year (mean age $\pm$ SD = 25.1 $\pm$ 6.9 year). Gestational age range from 29 to 40 weeks (mean gestational age $\pm$ SD = 35.6 $\pm$ 1.6 week).

Controls:

Sixty apparently healthy pregnant women attending the Antenatal clinic, and Labor Ward at Al-Kadhimiya Teaching Hospital, for re-evaluation of their pregnancy, or for delivery. The control groups were comparable preeclamptic groups regarding the age, gestational age, Depending on the gestational age, the apparently healthy pregnants were divided into two groups:

3**. Control pregnants in the second trimester (G3):**

They were thirty apparently healthy pregnants in the second trimester of pregnancy. Age range was from 15 to 38 years (mean age $\pm$ SD = 24.6 + 4.5 year). Gestational age range was from 20 to 28 weeks (mean gestational age $\pm$ SD = 25.5 $\pm$ 1.8 week).

4. Control pregnants during the third trimester (G4):

They were thirty pregnants in the third trimester of pregnancy. Age range was from 18 to 35 year (mean age $\pm$ SD = 24.8 $\pm$ 4.6 year). Gestational age range

was from 29 to 40 weeks (mean gestational age ± SD = 34.6 ± 2.1 week).

B. Blood & urine samples:

Ten milliliters of random venous blood were withdrawn from each patient and control, in supine position, without application of tourniquet. Samples then were transferred into clean new plane tube, left at room temperature for 15 minutes for clotting, centrifuged, and the separated serum was transferred into Eppendrof tube, which was used for measuring minerals (Ca, Mg). The tubes were stored at -20^{o} C until analysis, which was done within one month after collection [9].

Random urine specimens were obtained from each subject in the study to quantify urinary calcium [9], magnesium [9] that were expressed as a ratio to urinary creatinine [9]. As a preservative, 1-2 mls of 6M HCl was added to each random urine specimen; the samples were stored in appropriate containers at -20ºC until analysis [9].

C-Methods

Using atomic *absorption spectrophotometer* (Buck Scientific 210 JVP), the assay for calcium and magnesium estimation was carried out by adding 2.45 ml of (1% lanthanum chloride) solution to 0.05 ml of serum (or urine). These solutions were aspirated directly into air-acetylene flame where the calcium and magnesium hallow cathode lamp were used at wavelength 422.7 and 285.2 nm respectively [9]. *Adjusted serum calcium* can be calculated according to the formula [10]:

Adjusted calcium (mmol/L) = Measured calcium concentration (mmol/L) + 0.02 [40 – albumin concentration (g/L)].

Instead of obtaining a crude correction for measured calcium, the same data was used to calculate the ***ionized calcium*** according to the formula [10]:

Ionized calcium (mmol/L) =

$$\frac{60 \times \text{measured calcium (mmol/L)} - K'/12}{K' + 60}$$

Where

K' = 0.19 × total protein (g/L) + albumin (g/L).

The concentration of *magnesium ion* in serum was calculated from measurement of concentrations of total serum protein and total serum magnesium according to the equation [11]: $[100.4\ GZ / 100G - P]^2 + (33.77 + 2.42\ f\ P - f\ Mg)\ [100.4\ GZ / 100G - P] - 33.77\ f\ Mg = 0$. Where

P = total protein in gm per 100 ml of serum.

G = specific gravity of serum = 0, 00292 * P + 1.007

f = liters of serum that contain 1 kilo of water = 1000/G (4225.6 - 3225.6 G).

Mg = total magnesium in milli-equivalent per liter of serum.

Z = is Mg^{++} in milli-equivalent per liter of serum.

Results

The serum corrected and ionized calcium concentrations were lower in the preeclamptic women in third trimester G2 group as compared to healthy controls in the third trimester G4 [P < 0.001 for both] and even when compared to the preeclamptics in the second trimester G1 [P < 0.001 for both] as seen in (Table 1). The same significant reduction in *corrected but not ionized calcium* was noticed in the second trimester group G1 when compared to the healthy pregnants in the second trimester group G3 [P < 0.001 for corrected calcium but > 0.05 for ionized calcium] as seen in (Table 1). There was no significant difference in *corrected and ionized serum calcium* values between healthy pregnants in

each group [P > 0.05 for both] as seen in (Table 1).

Although the *urinary excretion of calcium (expressed as urinary calcium per creatinine)* was significantly reduced in preeclamptics in both groups G1 [P < 0.01] and G2 [P < 0.05] in comparison with pregnant controls of the same gestational period G3 and G4, the level of *urinary calcium excretion* was not significantly different between preeclamptics in the second and third trimester G1 and G2 [P > 0.05] nor between healthy pregnants in the same gestational periods G3 and G4 [P > 0.05] as seen in (Table 1).

A significant reduction in both *total and ionized serum magnesium* was noticed throughout the course of pregnancy whether among the preeclamptics groups: G1 and G2 [P < 0.001 for both]; or among healthy control pregnant G3 and G4 [P < 0.001 for both]. When preeclamptic groups G1 and G2 were compared with corresponding healthy control pregnant groups G3 and G4, the reduction in *total and ionized serum magnesium* was also significant [P < 0.001 for], as seen in (Table 1).

A significant elevation in *urinary magnesium excretion expressed as a ratio of urinary magnesium to urinary creatinine* was noticed through out the course of pregnancy whether among the preeclamptic groups: G1 and G2 [P < 0.001]; or among healthy control pregnant G3 and G4 [P < 0.01 for both]. When preeclamptic groups G1 and G2 were compared with corresponding healthy control pregnant groups G3 and G4, the significant increase in *magnesium excretion* is also found [P < 0.001], as seen in (Table 1).

A significant elevation in *the ratio between ionized calcium to ionized magnesium* was noticed in preeclamptics in both gestational period groups G1 [P < 0.001] and G2 [P < 0.001] as compared to the healthy pregnants in the same gestational periods G3 and G4. This significant elevation was also present in pregnants in the third trimester groups G2 [P < 0.01] and G4 [P < 0.001] when compared to pregnants in the second trimester groups G1 and G3, as seen in (Table 1).

Discussion

A number of studies have been published finding *serum total calcium* levels not different in non-pregnant controls and healthy pregnant women, whereas other researchers like Pederson et. al. [12], found decreased total serum calcium values in normal pregnancy. Furthermore, the beneficial role of a calcium supplementation in preeclampsia is still controversial [13, 14]. Some investigators reported an increased free erythrocyte and platelets calcium concentration, speculating that transmembrane calcium fluxes re-altered in hypertensive pregnancy, possibly by a specific mechanism probably of placental origin[7]. The finding of low serum total calcium in preeclamptics reported here is in agreement with findings of others [7, 15, 16] who conclude that a calcium deficit leading to an increased intracellular ionized calcium concentration during late pregnancy contribute to the pathogenesis of preeclampsia. In contrary, many investigators [6, 12, 17, 18] found that serum calcium did not differ significantly from normal pregnant group.

Regarding the ionized fraction of calcium, is crucial for the synthesis of vasoactive substances in the endothelium as prostacyclin and nitric oxide [19]. The finding of significant reduction in this

fraction, as seen in Table 1 is consistent with those reported by Seely et.al. [20], who revealed that a low level of active vitamin D (1, 25-$(OH)_2$ D) in preeclamptics, may contribute to suboptimal intestinal absorption of calcium during a time of increased calcium demand resulting in lower ionized calcium, increased PTH, and hypocalciuria in preeclampsia[6]. Abnormalities in calcium homeostasis may contribute to the increased vascular sensitivity documented in preeclampsia [6]. In contradiction to the reported difference in ionized calcium between normal and preeclamptic patients, other authors like Sanders et.al.[17], Siddiqui & Rana[21], Pederson et.al.[12], Richards et. al.[22] found no difference in serum ionized calcium.

Urinary calcium in preeclamptic in this study was observed to be lowered as compared to corresponding control pregnant as seen in (Table 1).

The etiology of hypocalciuria in preeclampsia is unknown. However, different assumptions have been given [23]. Particularly, it has been proposed that hypocalciuria may result from decreased dietary intake of calcium resulting in a low circuating calcium and hence low urinary calcium[23]; or from decrease intestinal absorption as secondary result of decreased 1,25 dihydroxyvitamin D, which enhances intestinal absorption of calcium[23]; or it may be due to increased calcium intake by the growing fetus and placenta[23]; lastly, it may be due to intrinsic renal tubular dysfunction, presumably due to decreased glomerular filtration and increased tubular reabsorption[23].

We found also, a decrease in both total and ionized magnesium throughout the 2nd and 3rd trimesters of pregnancy in both normal and preeclamptic pregnant women as seen in Table 1, like several studies [6, 7, 24-28]. The level of the cation studied was found to be within the same ranges reported for corresponding non-pregnants in other studies like [7, 20, 24, 27]. Although the reason for the reduction in total and ionized magnesium is not clear, it is not likely to be due solely to hemodiluton and extracellular fluid volume expansion as serum magnesium levels are still observed to decrease when correcting for protein dilution[6]. An increase in the renal clearance during pregnancy may contribute to the reduction in serum magnesium, since the kidney is the main regulator of the body magnesium [6]. This was supported by the finding of significant increase in magnesium excretion in healthy control and preeclamptic pregnancy with advancing gestational age according to magnesium: creatinine ratio, as seen in (Table 1). Other factors that may contribute to hypomagnesaemia in pregnancy include poor dietary intake[6] which is accompanied by consumption of minerals by the growing fetal skeletal system [6]. Hypoproteinaemia is another contributing factor since extracellular magnesium accounts for about 1% of the total body magnesium content. About 55% of magnesium is free, 30% is associated with proteins (primarily albumin), and 15% is complexed with phosphate, citrate, and other anions [9]. The technique used for measuring ionized magnesium can also be considered, ideally, it is the ion-selective electrode which is not available in Iraqi laboratories, instead a mathematical equation was employed [9, 11].

We also found an increased ionized calcium:ionized magnesium ratio during normal and complicated pregnancy, as seen in (Table 1). In

previous reports [28], the molar ratio of total calcium to total magnesium remained constant throughout pregnancy. However, ionized magnesium can be altered independent of total magnesium concentrations [29]. A high calcium-magnesium ratio has been associated with increasing vasospasm [30]. Increased intracellular calcium and decreased intracellular magnesium have been reported in women with hypertension and diabetes [30]. Thus electrolytes abnormalities may contribute to altered blood pressure [23].

The relation between serum total and ionized magnesium with intracellular magnesium has not been defined clearly. In previous study [30], there was no significant difference in red blood cell magnesium levels in teenagers with pregnancy-induced hypertension, whereas plasma magnesium tended to decrease with increasing gestation in this same group. However, recent evidence suggests that extracellular magnesium may modulate intracellular magnesium in vascular smooth-muscle cells [6].

On the basis of previous experimental data, the mechanisms underlying the *magnesium-induced vasodilation* may be due to a modification of the response to vasopressor hormones [7], and an interaction with cellular calcium handling[7]. These possible mechanisms were discussed by Kisters et. al. 2000 [7].

Further study of intracellular minerals and the membrane Na, K ATPase and calcium pumps to explore their potential role in the pathogenesis of preeclampsia is required for future work.

Table 1: The mean value of minerals (corrected Ca^{+2}, ionized Ca^{+2}, total Mg^{+2}, ionized Mg^{+2}, ratio of ionized Ca^{+2}: ionized Mg^{+2}) in the sera & urine of different preeclamptic and pregnant control groups (presented as mean ± SD).

Variable	G1	G2	G3	G4
Serum corrected calcium (mmol/L)	2.3 ± 0.05	2.2 ± 0.09	2.5 ± 0.1	2.5 ± 0.1
Serum ionized calcium (mmol/L)	1.2 ± 0.08	1.1 ± 0.05	1.2 ± 0.05	1.2 ± 0.05
Urinary calcium : creatinine	0.6 ± 0.27	0.58 ± 0.59	0.94 ± 0.6	0.8 ± 0.19
Serum magnesium (mmol/L)	0.11 ± 0.005	0.08 ± 0.01	0.15 ± 0.007	0.13 ± 0.0006
Serum ionized magnesium (mmol/L)	0.011 ± 0.002	0.006 ± 0.001	0.057 ± 0.0037	0.018 ± 0.0010
Urinary magnesium: creatinine	0.07 ± 0.003	0.09 ± 0.03	0.0149 ± 0.0101	0.04 ± 0.01
Serum ionized calcium: ionized magnesium ratio	172.37 ± 36.36	250.64 ± 134.32	32.25 ± 2.45	101.06 ± 7.1

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Extraction and purification of two outer membrane proteins (porins) from *Klebsiella pneumoniae* local isolate.

Amir H. Al–Shammary[1] *PhD*, Essam F. Al-Jumaily[2]*PhD*, Nidhal Abdulmohymen[1] *PhD*.

Abstract

Background: The porins are present in large amounts in the outer membrane of gram negative bacteria and form water-filled channels that permit the diffusion of small hydrophilic solutes across the outer membrane. Porins are generally divided into two classes: nonspecific porins (e.g., OmpC and OmpF), which permit the general diffusion of small polar molecules (600 Da), and specific porins (e.g., LamB), which facilitate the diffusion of specific substrates.

Objective: To purify and characterize outer membrane proteins (porins) from a local isolate of *Klebsiella pneumoniae*.

Materials and methods: An identified local isolate of *Klebsiella pneumoniae* was used as a primary source for the isolation and purification of porins. Outer membrane protein (porins) was purified and characterized and the contaminating lipopolysaccharides (LPS) were detected by thiobarbituric acid assay.

Results: The final preparation contained porins in a concentration of 3.2 mg/ml. The results of electrophoretic separation revealed that porins appeared as two distinct bands with molecular weights of porins were estimated to be 35 and 36 kDa, respectively.

Conclusions: Porins were expressed by the local isolate of *Klebsiella pneumoniae* with molecular weights highly similar to that of porins preparations produced by other gram negative bacteria and *Klebsiella pneumoniae* expressed two types of porins under standard laboratory conditions.

Keywords: Porins, Thiobarbituric acid, Gel filtration chromatography, Ketodeoxyoctinate.

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Introduction

Approximately, 50% of the dry mass of the outer membrane of gram-negative bacteria consists of proteins, and more than 20 immunochemically distinct proteins (termed outer membrane proteins [OMPs]) have been identified in *E. coli*. Apart from their structural role, OMPs have also been shown to have other functions, particularly with regard to transport, and have been classified as permeases and porins. Furthermore, several OMPs have been shown to be potent inducers of cytokine synthesis [(1)].

Porins are OMPs which form trimers that span the outer membrane and contain a central pore with a diameter of about 1 nm. These porins (e.g., OmpC and OmpF of *E. coli*) are permeable to molecules with molecular masses lower than approximately 600 Da. Porins play a crucial role in the interactions between the environment and bacteria, in addition, or probably as a consequence, they are present in large amounts in the outer membrane of gram-negative bacteria [(2)].

Materials and methods

Porins were extracted according to the method described by Nurminen [(3)]. Briefly, the bacterial cells were harvested by centrifugation at 4000 rpm for 30 minutes. One gm of bacterial cells was washed twice with 0.01M tris buffer (pH 7.8) and suspended in10 ml of 0.01 M tris

[1]Dept. Medical Microbiology, College of Medicine, Al-Nahrain University, [2] Institute of Genetic Engineering and Biotechnology for postgraduate studies.
Adress Correspondence to: Dr. Amir H. Al–Shammary
E- mail: amer_hani@yahoo.com
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buffer containing 0.01 M EDTA and 1.3 mg lysozyme, then 0.4 ml solution of 1 M $MgCl_2$ containing 50 mg DNas & RNase each was added. One gm (wet weight) prepared above was extracted twice (separated by centrifugation at 3000xg) with 2% TX-100 buffer containing 0.01M $MgCl_2$.

Half mg trypsin / ml suspension was added and incubated overnight at 37 °C. One hundred ml of the digested mixture was centrifuged at 20000xg, the supernatant was collected, and the pellet was digested once more with trypsin. The supernatant of both digestions was ultrafiltered using the amikon apparatus. The retained material was washed with 1L of D.W. by further ultra filtration. The precipitate was suspended into 100 ml of D.W. and centrifuged at 20000xg for 20 minutes. The sediment was finally suspended in 10 ml of D.W.

For further purifying porins, the final preparation was subjected to gel filtration chromatography using Sephacryl S-200 gel.

Preparation and packing of the gel

Sephacryl S-200 gel was prepared according to the instructions of the manufacturing company. It was suspended for 2 hrs in 250 ml of 0.01 M EDTA buffer (pH 7.5) containing 0.2% TX-100 and then it was degassed by using vacuum pump. Gel was poured with care (to avoid bubbles) onto a column with dimensions of 1.5x88 cm. Finally the column was equilibrated over night with the same buffer.

Method of Gel filtration chromatography

Five ml of porins solution was loaded onto the column, and fraction of 5 ml each were eluted after settling the flow rate to about 30 ml/hrs. Absorbance at 280 nm was measured for all of the fractions.

Concentration

The porins peaks, were collected as 80 ml of elution buffer and concentrated by sucrose to a final volume of 10 ml for each peak.

Sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE)

The purity of the porins and the apparent masses of their variants were estimated by SDS-PAGE. SDS-PAGE was done according to the method of Laemmli described by Garfin, [4]. The protein concentration in the final preparation of porins was measured by the absolute method:

$$\frac{\text{O.D. at 235nm - O.D. at 280 nm}}{2.51} = \text{protein concentration (mg/ml)}$$

as mentioned by Whitaker and Granum, [5].

Thiobarbituric acid assay for the estimation of lipopolysaccharide (LPS) concentration (Ketodeoxyoctinate).

Standard curve of LPS: Several known concentrations of LPS were plotted versus their relevant absorbance at 550 nm, and a standard curve was created. By the aid of the standard curve, it was possible to measure LPS concentrations in the final porins preparation.

Thiobarbituric acid assay was performed according to the method described by Hanson and Philip, [6] and to alleviate the cytotoxic effects of contaminating LPS, polymyxin B was added to the final porins solution in a dose of 5 g/ml and the mixture was incubated for one hour at 20°C.

Results

The results revealed that porins were eluted as two peaks (Figure 1); the fractions enriched in protein, identified by absorbance at 280 nm, were pooled and extensively concentrated by sucrose and checked

for protein heterogeneity by SDS-PAGE. Figure 2 illustrates the electrophoretic experiment of the present study which shows clearly that porins are represented by two bands. The first is at molecular weight 36 kDa and the other at molecular weight 35 kDa. The two bands are related to peak no. 1 and peak no. 2 shown in figure 1, respectively. The final preparation resulted after purification steps contained porins in a concentration of 3.2 mg/ml as estimated by the absolute method and it was shown that porins solution had contaminating LPS in a concentration of 117 µg / ml (Figure 3).

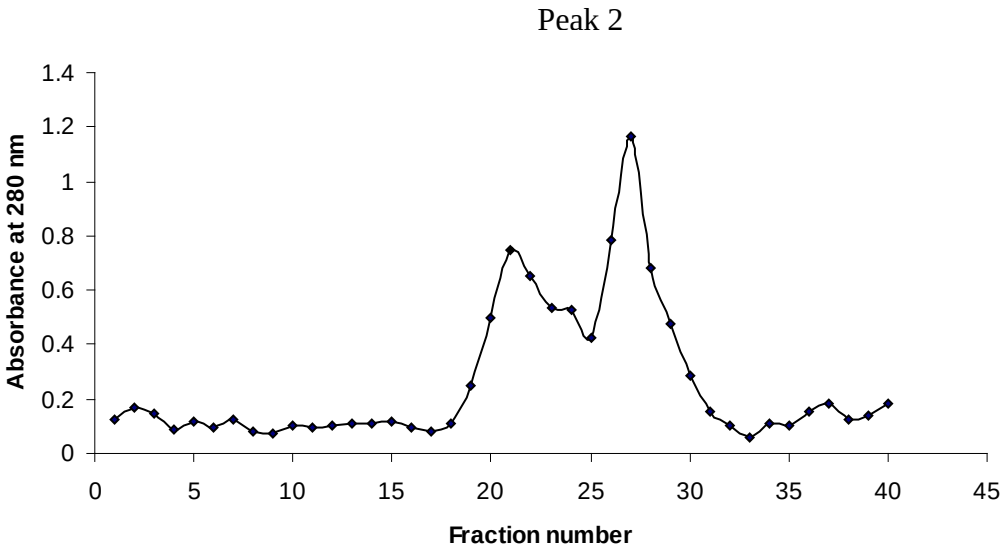

Figure 1: Purification of *Klebsiella pneumoniae* porins with Sephacryl S-200. The dimensions of the column was 1.5x88 cm, flow rate was adjusted to 30 minutes/hrs. 0.2% TX-100 (pH 7.8) containing 0.01 M EDTA was used as elution buffer and fraction of 5 ml each were eluted.

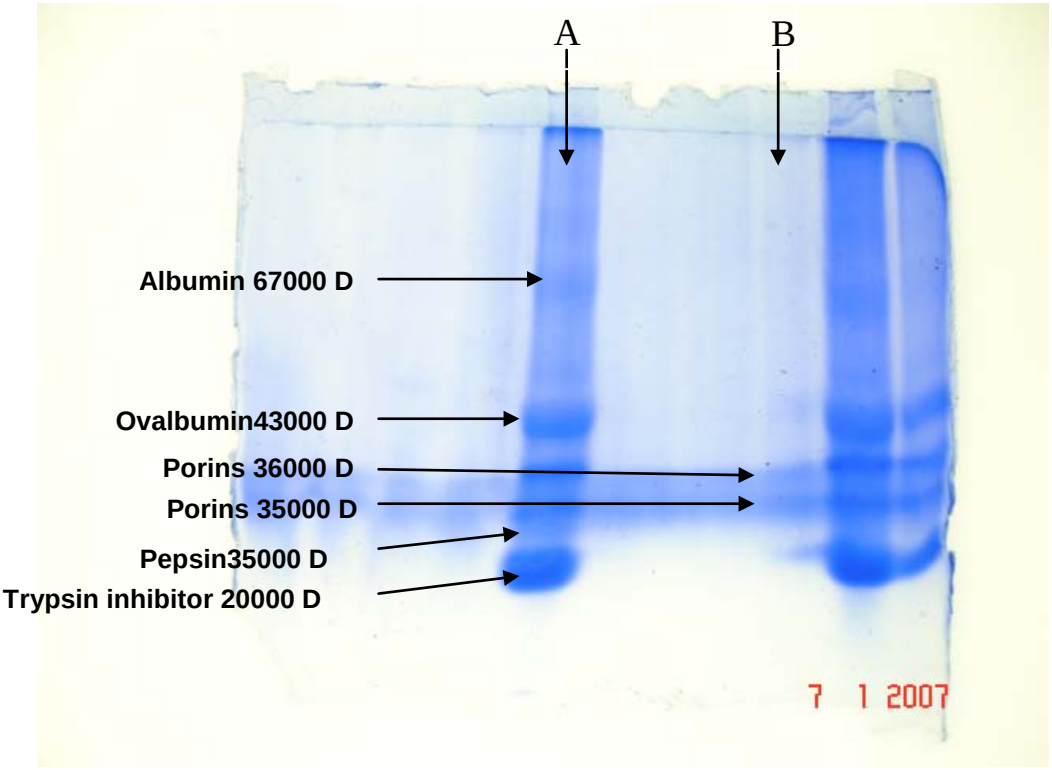

Figure 2: SDS polyacrylamide gel electrophoresis of porins.
A. Standard proteins.
B. Porins sample.

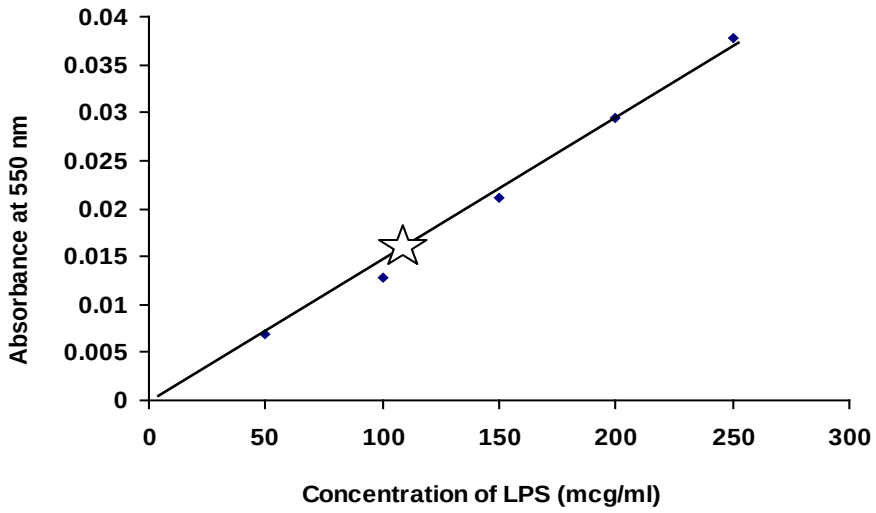

Figure 3: Standard curve of Lipopolysaccharides (LPS)

Discussion

Porins have been purified from a number of gram negative bacteria. In all so far examined cases; the apparent molecular weights of the proteins are in the range of 30 to 45 kDa, while some of the porins can be purified as oligomers in SDS. They are usually acidic proteins and in many cases show association with peptidoglycan. In addition, outer membrane contains multiple species of porins. The separation of one porin from the other is difficult, for example, one can suppress the production of one or more porins through manipulation of culture conditions [(7)].

Among the outer membrane proteins found in gram-negative bacteria are the abundant porins which form diffusion channels for small molecules such as metabolizable sugars [(8)]

It is well documented that disruption of cells will increase membrane protein yield. Thus, enzyme digestion was used to disrupt the bacterial cells, and the use of lysozyme – EDTA greatly enhances membrane destruction [(9)]. Furthermore, the use of DNase and RNase would result in degradation of nucleic acids and increasing the purity of proteins [(10)].

The protease resistance capability of the porins could be utilized for their isolation. For example, trypsine solubilizes practically all the proteins of the TX- treated envelopes degrading at the same time most of them with the exception of porins [(3)].

The results of this study are expectedly consistent with the results of Galdiero & Co-workers, (1994) [(11)] and Meghji &Co-workers, (1997) [(8)] who mentioned that the purified porins from *Salmonella typhimurium* showed the two expected bands with molecular masses of 34 and 36 kDa and the purified porin from *Pseudomonas aeruginosa* showed two bands with a molecular mass of 36 to 38 kDa, respectively. The apparent similarity of the results might indicate phyllogenetic relationship since outer membrane proteins are conserved, with minor differences, in all members of the gram negative bacteria.

Porins possess a high proportion of β-sheet structure, which traverses the membrane in a tightly packed β - barrel organization. This makes them relatively resistant to denaturation by SDS or other detergents at low temperature but not at higher temperatures [(12)]. Therefore, porins display different motilities when they were separated at low or high temperatures [(13)].

Porins usually have a strong association with LPS and it is difficult to obtain the proteins completely free of LPS contamination [(14)].

It is stated that, by binding to the lipid A of LPS, polymyxin B completely inhibits the strong cytopathic effect of this lipid whereas binding to the porins leaves the biological activity of the protein unmodified [(15)].

It is stated that a concentration of 0.5-1.0 mg/ml of LPS is required for *in vitro* cytotoxic effect [(16)]. The results of the current study revealed close proximity to that of Luo & Co-workers, (1997) [(17)] when they estimated LPS concentration of 0.418 µg of Ketodeoxyoctinate (KDO) / mg of protein. While in another study, it is mentioned that the content of LPS in porins preparations was in the order of 1 pg/mg of porin [(7)]. Furthermore, in another experiment to extract and purify porins from *Pseudomonas aeruginosa*, much lower value for LPS was recorded in the final preparation (about 20 µg/ ml) which was neutralized by incubation with polymyxin B as mentioned above [(15)].

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Evaluation of the role of erythrocyte deformation on erythrocytes aggregation and sedimentation rate using He-Ne laser scattering

Rowaida A. Al-khazragi *MSc.*

Abstract

Background: The erythrocyte aggregation is an important physiological phenomenon in the circulation of blood. It is a basic characteristic of normal blood that plays a major role in the cardiovascular system, especialy in the microcirculation.

Objective: To evaluate the role of deformability of red blood cells on the aggregation and sedimentation of red blood cells.

Subjects & Method: The present study was carried out on thirty two healthy subjects. Laser scattering method was employed for this study. From scattered light intensity, profiles continuously obtained during aggregation and sedimentation of the aggregated erythrocytes. Different values of erythrocyte deformability were determined and evaluate their effects on each phase of the erythrocyte aggregation and sedimentation, rouleaux formation, one-dimensional aggregate and three- dimensional aggregate formation.

Results: Deformability values are expressed in term of rigidity index, the difference between medium and high rigidity index significantly decreased the rate of aggregation and the rate of three dimensional aggregate sedimentation.

Conclusion: Variation of the values of erythrocyte deformability from low to medium and from medium to high showed different effects on aggregation and sedimentation stages.

Keywords: Erythrocyte aggregation, sedimentation rate, deformability, laser light.

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Introduction

Erythrocytes aggregation and disaggregation are natural phenomena in the circulation of blood [(1)]. Aggregation of red blood cells is the formation of reversible structure containing a number of particles, while erythrocytes sedimentation monitors the tendency of red blood cells to form aggregates in plasma [(2)].

The formation of clumps of red blood cells under low or non-flow conditions, result from the attraction forces between the red blood cells. The cells adhere to each other in rouleaux aggregates. Slight mechanical force, such that occurs in the circulation, is enough to disperse these aggregates.

The process of aggregation affected by many physical and chemical factors. Chemical factors are concerned with modifications of either erythrocytes or suspending medium such as hematocrite, PH of the suspending medium, macromolecules and flow conditions [(3,4)].

Some investigations have pointed out the importance of cellular modifications to the erythrocyte aggregation, especially in relation to erythrocyte deformability, and erythrocyte filterability, (Ability of erythrocytes to change shape as they pass through narrow spaces, such as the microvasculare) [(5)].

Dept. Physiology, College of Medicine, AL-Nahrain University.
Adress Correspondence to: Dr. Rowaida A. Al-khazragi .
E- mail: Rowada_AbdulAmeer@yahoo.com
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Figure 1: Erythrocyte deformability when passing through microcirculation

In large blood vessels, the resistance to blood flow depends to minor degree on blood viscosity but mostly upon the diameter of the vessels. This is due to the laminar blood flow and the deformability of erythrocytes under the high shear rate which reduce the viscosity of blood makes it ineffective. While the resistance to blood flow in the capillaries depends mainly on viscosity of the blood and erythrocyte aggregation under the low shear rate which increase blood viscosity [(6)].

The true capillaries are about 5 µm in diameter at the arterial end and 9 µm at the venous end, and since the red blood cells are flat disks of about 7 µm in diameter, thus when the sphincters are dilated, the diameter of the capillaries is just sufficient to permit erythrocyte to squeeze through in a "single file". Erythrocyte is a "bag" that can be deformed into almost any shape. This is because the normal cell has a great excess of cell membrane for the quantity of material inside, and due to the pliability of the cell membrane. Deformation does not stretch the membrane greatly & consequently does not rupture the cells as would be the case with old erythrocytes [(6,7,8)].

Materials and methods

The present study depend on a modified method of E. Muralidharan, in Biorheology, 1994,[(9)].Which work on the same principle of laser light scattering.

Five ml of fresh blood samples were drawn from the cubital vein of 32 healthy human subjects using heparin (0.03/5ml of blood), as anticoagulant, into a sterilized tube. 1 ml of the blood was used to measure the ESR (erythrocyte sedimentation rate) by Westergren method. Any record above 15 mm/hr for males and above 20 mm/hr for females) was excluded from the study (According to Bottlger, 1967) [(10)].

Four ml of the rest blood sample was put in centrifuge to separate erythrocytes from plasma and WBC coat

(3000 rpm for 10 minutes at 4°C). After removal of plasma & WBC coat, the erythrocytes were washed 3 times with iced cold normal saline (0.9% NaCl) and then re-centrifuge the sample again (3000 rpm for 10 minutes at 4 °C) to remove any particles that may be attached to the erythrocyte membrane.

Physiological measurements:

The measurement of erythrocyte deformability:

It was carried out by measuring the filtration time of the following two solutions:

The first solution 2 ml consisted of 5% of packed erythrocytes suspension prepared by mixing 100 µl of packed erythrocytes with 1900 µl of suspending medium.

(The packed erythrocytes were already washed three times with ice cold 0.9% NaCl as mentioned above).

The second solution (2 ml) was of cell free suspending medium.

The suspending medium was prepared from the following chemicals: (150 mM KCl, 0.5 mM Na_3EDTA, 10 mM Tris-HCl)

The pH of the solution was 7.4 at room temperature.

The filtration was through Whatman filter paper (number 1).

The deformability values were expressed in term of rigidity index (RI):

$$RI = \frac{\text{Filtration time of 2 ml 5\% packed erythrocytes suspension}}{\text{Filtration time of 2 ml of cell free suspending medium}}$$

This method was described by Al-Gailani and Al- Remadani (1998) [11].

The blood sample preparation:

To prepare the sample, 800 µl of dextran (mol. Wt.500.000) was added to phosphate buffered saline (P.B.S.) solution (50mM Sodium Phosphate, 3mM KCl, 90mM NaCl, 0.1g/dl D-glucose, PH 7.4), then 100 µl of bovine serum albumin of 0.5g/dl concentration was added to the tube, to prevent adhesion between RBCs and the wall of chamber, followed by addition of 100 µl of packed erythrocytes to get, after mixing, sample of 10% PCV. The experiments were carried out at room temperature.

The system of the measurement is shown in Figure 2; it is compose of a linear polarized He- Ne laser source, of wave length 632.nm, (Griffin Co.), and generation power 1mW. A beam of 1mm diameter was passed through erythrocyte suspension in a chamber of 50×10×1 mm; made of a microscopic glass plates, figure 3. Blood column hight was kept at 40mm. The forward scattered light intensity through the sample column was detected with a photocell (photodiode ampliphier) [9].

The photocell placed in front of the laser beam and allowed the beam to pass directly through the crystal of the cell. The signals from the photocell are passed through a light flexible cable to an amplifier (Grass 7P1F) for signal amplification. The sample chamber was mounted firmly on the holder so that the laser beam passed, exactly, through the center of the chamber.

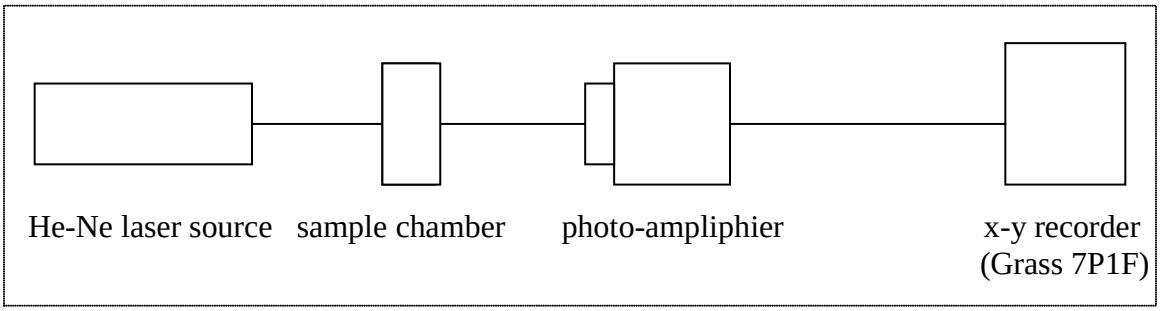

Figure.2: The system layout

The blood sample was gently introduced into the chamber by using a syring with long needle. Immediately after the sample was introduced, the forward- light signal was continuously recorded by the system.

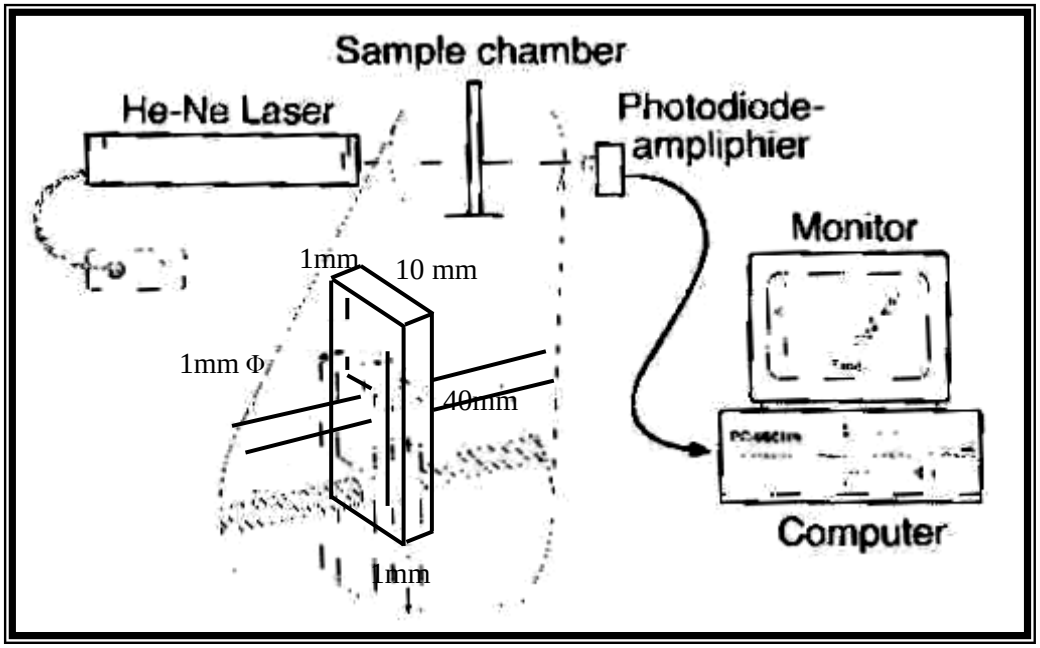

Figure. 3: Schematic diagram of an arrangement of light scattering experimental system (Muralidharan, 1994)

Results

Figure 4 shows the pattern of rouleaux formation, one-dimensional aggregate and three- dimensional aggregate formation curve, with sample of 10%PCV as it recorded by laser assessed aggregometry used in this study.

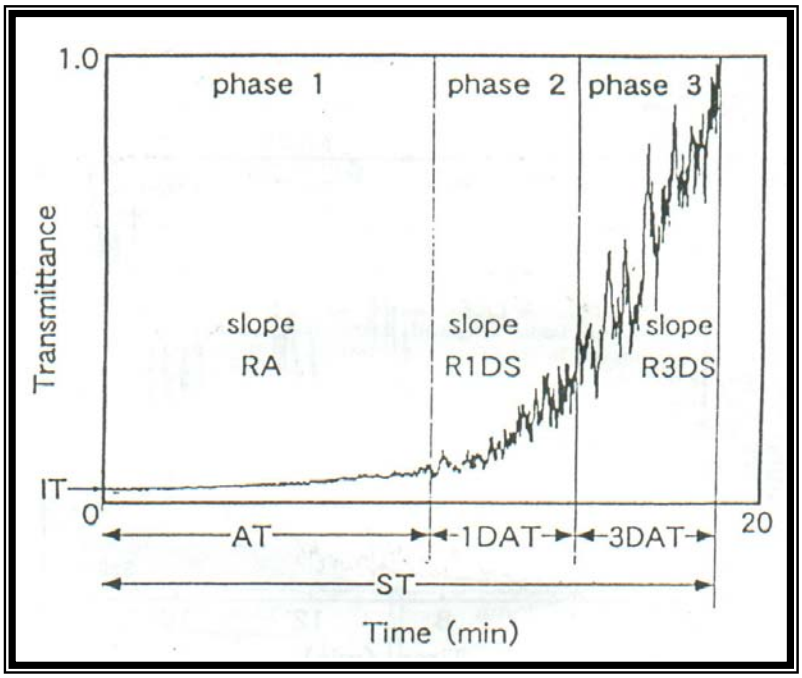

Figure 4: Pattern of different stages of aggregation and sedimentation as recorded by laser scattering techniques.

There was a slight increase in the signal due to the reorientation of single erythrocytes when the erythrocytes were monodispersed in the beginning of the aggregation process. The sedimentation of the aggregates formed was indicated by the appearance of fluctuations in the signal. These fluctuations were smaller in the beginning and became larger towards the end. The time at which the first sharp fluctuation appeared in the signal was termed AT (aggregation time). These fluctuations continued until the signal reached the maximum without any variation. The time at which the signal reached the maximum was termed ST (sedimentation time). The initial phase was due to the movement of single erythrocytes in the process of forming small aggregates. The rate of aggregation (RA) was obtained from the slope of this phase. The second phase was due to the sedimentation of small and one-dimensional aggregates. The duration of this phase was termed 1DAT (one-dimensional aggregation time).

The slope of this phase provided the rate of sedimentation of one-dimensional aggregates (R1DS). The third phase was due to the sedimentation of large and three- dimensional aggregates. The duration of this phase was termed 3DAT (three-dimensional aggregation time).

The rate of sedimentation of the three-dimensional aggregates (R3DS) was obtained from the slope of this phase. The light intensity fluctuation showed a clear visible in the signal between these phases.

Table 1: The values of ESR and deformability in this study

	ESR (mm / hr)	**Deformability (RI)***
Mean	**3.11**	**1.59**
SD (±)	**2.96**	**0.23**
Max.	**12**	**2**
Min	**1**	**1.25**

* Deformability expressed in term of rigidity index

The deformability of erythrocytes in this study was expressed in term of rigidity index, their values was ranged from 1.25 to 2 with a mean of 1.59±0.23.

The rigidity index results had been classified into 3 groups: low (1.25-1.49), medium (1.5-1.74) and high (1.75-2.0) as shown in table 2.

Table 2: Effect of low, medium and high erythrocyte deformability on aggregation and sedimentation parameters

		Low n=12	Medium n=11	High n=9
Time	AT	13.82±2.71	13.51±3.33	12.99±2.78
	1DAT	6.27±3.38	6.23±3.9	5.31±1.49*
	3DAT	2.03±1.06	2.76±1.3	2.47±0.76
	ST	23.02±4.55	22.49±6.86	20.77±3.27*
Rate	RA	0.037±0.008	0.039±0.007	0.045±0.015*
	R1DS	1.39±0.59	1.71±0.7	1.76±0.41
	R3DS	9.87±3.95	9.86±3.96	9.73±2.01*

* Significant difference between medium and high groups of erythrocyte deformability.

Table 2 shows a significant decrease ($P<0.01$) in the sedimentation time ST, one dimensional aggregation time 1DAT and in the rate of sedimentation of the three dimensional aggregates R3DS at high rigidity indexes of erythrocytes.

On the other hand there was a significant increase ($P<0.05$) in the rate of

aggregation RT at high rigidity indexes of erythrocytes.

Discussion

Some investigators have pointed out the importance of cellular modifications to the erythrocyte aggregation especially in relation to erythrocyte deformability. The contributions of the cellular alterations to the erythrocyte aggregation are smaller in magnitude, but the influence is significant [(5)].

The major determinants of the ability of deformation (deformability) of erythrocyte are enhanced by: [(12)]

a. Extracellular viscosity.
b. Membrane stiffness.
c. Cell geometry (surface area /volume ratio).

In the present study the deformability expressed in term of rigidity index (membrane stiffness) of erythrocyte.

When we compared the deformability results (RI) in this study with the aggregation stages and sedimentation, there was a significant decrease in the time needed for one dimensional aggregate formation (1DAT) and for sedimentation and significant increase in the rate of aggregation at high (RI) of erythrocyte, this is due to the presence of the macromolecules, which play the main role in the first stage of the aggregation process. The macromolecules act as a bridge and permit the RBCs to slide on each other to form rouleaux in the suspending medium [(13, 14)].

On the other hand, there was a significant decrease in the rate of the third stage (3DAS) at high (RI) of erythrocyte, this is due to decreased the deformability of erythrocytes, because the third stage of aggregation process depend on the shape and the deformability of erythrocytes and decreased the erythrocytes deformability made the size of the aggregate formed by erythrocytes of high (RI) are small, and the erythrocytes in this stage (three dimensional aggregates) are loosely packed [(15,16)].

This study showed different values of deformability have causes different effects on the three stages of aggregation and in turn on sedimentation.

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Cystinuria in a group of children in Iraq

Shatha Hussain Ali *CABP*.

Abstract

Background: Cystinuria is an autosomal recessive defect in reabsorptive transport of Cystine and dibasic amino acids. Increased urinary excretion of Cystine, the least soluble of all amino acids, results in formation of stones**.**

Objectives: we report our experience with management of cystinuria in a group of Iraqi children.

Patients and Methods: from 1999 to 2006, all children with cystinuria were evaluated, treated and followed in Al – Kadhimiya Teaching Hospital.

Results: Twenty three patients with cystinuria having calculi (16 males, 7 females) were treated. Their age ranged from 10 months to 18 years.

Associated hyperuricosuria was recorded in 30.5%, hypercalciuria in 13% and hyperoxaluria in 4.3%. Follow up period ranged from 1 – 88 months.

Nine patients were treated with increased oral fluids and alkalis only.

D–Penicillamine therapy was given to 13 patients. Side effect to penicillamine was noticed in 4 patients (22.2%). Captopril was given to 4 patients. Extracorporeal shock wave lithotripsy (ESWL) was performed in 8 patients, and 18 patients underwent open surgical procedures.

The stone free rate was 55.6% with fluids and alkali alone, 58.3% with D–Penicillamine, 0% with Captopril and 50% with ESWL.

Combined treatments were required in 45% of patients. Stone recurrence rate was 70%.

Conclusion: Oral fluids and alkali was most successful when used in patients with mild disease. D–Penicillamine and ESWL had nearly equal rate of successful results.

Keywords: cystinuria, chlidren, calculi, urolithiasis

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Introduction

Cystinuria is an autosomal recessive defect in reabsorptive transport of Cystine and dibasic amino acids: Ornithine, Arginie and Lysine from the luminal fluid of the renal proximal tubules and small intestine [1 – 10].

Increased urinary excretion of Cystine (the least soluble of all amino acids) results in Cystine crystallization and formation of stones [1 – 10].

Cystinuria is the cause of 1 – 2% of stones observed in adults and up to 10% of those occurring in children [1, 3, 4, 8].

Dept. Pediatrics,College of Medicine, Al–Nahrain University.
Adress Correspondence to: Dr. Shatha Hussain Ali.
P. O. Box 70074, Mobile: + 964 07901479929
E-mail: shathah666@yahoo.com
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In 1966, Rosenberg et al, described three types of Cystinuria according to urinary phenotype: I, II, III [1 – 5, 8, 10].

A new classification of the disease by the International Cystinuria Consortium follows the chromosomal localization of the mutation with type an on chromosome 2, type B on chromosome 19 and type AB on both chromosomes [1, 4, 8].

The only phenotypic manifestation of Cystinuria is Cystine urolithiasis, begins in the first decades and often recurs throughout a patient's life time [1, 3, 4, 10]. Surgical intervention is often necessary, but the cornerstones of treatment is medical prevention of recurrent stone formation [1, 3, 4, 8, 10].

Aim of study

To report our experience with presentation, management, clinical

course and outcome of a group of Iraqi children with Cystine urolithiasis.

Patients and methods

This prospective study was conducted in the Pediatric Nephrology Clinic in Al - Kadhyimia Teaching Hospital in Baghdad between January 1999 and April 2006.

Twenty-three children with Cystinuria, were evaluated, treated and followed.

Clerking included recording age, gender, age of onset of symptoms, age of diagnosis of stone disease, clinical presentation, past medical and surgical history, recurrence and family history of stone disease.

Every child was tested for plasma urea, creatinine, sodium, potassium, calcium, phosphorous, uric acid and alkaline phosphatase level. Their urine was analyzed by microscopy and cultured.

Twenty four hour urinary determination of calcium, oxalate and uric acid was done to all children. They remained on their usual diet. In the morning, their first specimen of urine was discarded; urine was then collected for 24 hours. In young children who are not toilet trained, urine was collected by use of adhesive urine bags. For older children, collection was into container.

Hypercalciuria (HCa) was defined as urine Calcium excretion of > 4 mg/ Kg/ 24 hr. [6, 11, 12, 13]. Hyperoxaluria (HOx) was defined as urine Oxalate excretion of > 55 mg/ 1.73 m^2/ 24 hr [6, 12, 13]. Hyperuricosuria (HUr) was defined as Uric acid excretion of > 815 mg/ 1.73 m^2/ 24 [6, 12, 13]. Urine amino acid excretion was tested in all children using thin layer chromatography as well as Cyanide – Nitroprusside test [5, 14].

Stones, when available, were analyzed chemically.

Patients were diagnosed as having Cystinuria on the basis of one or more of the following criteria:

1. Cystine crystalluria: was observed in 2 patients (18.7%) on urinalysis [1, 4, 5, 8, 10, 12].

2. Positive Cyanide – Nitroprusside test in all patients. [1, 4, 5, 8, 9, 12, 15].

3. Detection of urinary excretion of amino acids Cystine, Ornithine, Arginine and Lysine by thin layer Chromatography in all patients [5, 10, 12, 14].

4. Cystine (pure or mixed) composition of calculi obtained from 10 patients by chemical analysis [1, 4, 5].

All stones were documented radiologically by Renal Ultrasonography (US) and Intravenous Pyelography (IVP). Voiding Cystourethrography (VCUG) was done when Vesicoureteric Reflux (VUR) was suspected.

Treatment Programs included:

1. Oral fluids and Alkali: All patients were instructed to maintain a fluid intake throughout day and night to ensure at least 1.2 L – 2 L / m^2 or 40 ml/ Kg/ day of urine daily. The urine should be kept very light in color approaching that of tap water making it a habit to observe this aspect with each voiding [16]. Adequate hydration was monitored on follow up visits every 1-2 months by checking the specific gravity achieving 1.010 [1].

2.Alkalinization of urine to a PH at about 7.5 throughout the day and night was accomplished with oral administration of alkali in the form of Potassium Citrate 60 – 80 Meq/ day or Sodium Bicarbonate 0.2 g/ Kg/ day in divided doses. Dose was adapted to urinary pH. Checking urine PH was done by the patients at home using urine strip and in laboratory on follow up visits. Moderate salt intake was

instructed to all patients, not to add salt to their diet [1, 3, 4, 8, 9, 10, 12]. Limit intake to 50 mmol/ day [3].

3. D– Penicillamine (DP). : Starting dose of 20 mg/ Kg/ day was increased gradually up to 40 mg/ Kg/ day given orally in 2 - 4 divided doses with half the dose given at bed time [1, 3, 4, 8, 12]. Most of the patients received 3 divided doses

4. Captopril: Given orally in a dose of 2 mg/ Kg/ day with observation of blood pressure [1, 3, 4]. The patients were given 2 divided doses.

5. Extracorporeal shock wave lithotripsy (ESWL): ESWL was performed using the Siemens Lithostar lithotripter. Youngest patient among our series was 12 month of age.

6. Open Surgical Procedures done with cooperation of urology surgeons.

Outcome of various treatment modalities were categorized as: stone free, no change, decreased size and increased stone size [5] .

Results

Total number of patients was 23. Males were 16, females were 7. Their age ranged from 10 months to 18 years (median = 46 months and mean of 69.7 ± 12.8 months). The age at onset of first symptom attributable to stone disease ranged from 2 to 132 months (median = 17 months and mean of 35.1 ± 7.9 months). Age at the time of diagnosis of Cystinuria ranged from 2 to 132 months (median = 17 months and mean of 38.6 ± 8 months). Sixteen patients were diagnosed as cystinuria at their initial clinical presentation. Majority of patients (78.3%) had the onset of stone disease before 5 years of age. Sixteen patients (69.6%) recalled a family history of stone disease, 12 of 16 (75%) had parental consanguinity. The clinical presentation is shown in table 1. Most patients presented with passing stones spontaneously (52.2%) and gross hematuria (47.8%). Four patients presented with failure to thrive (FTT), one of them had associated celiac disease, the 2nd had recurrent UTI, the 3rd catch up growth on follow up and he was stone free and the 4th was lost to follow. Associated culture proven Urinary Tract Infection (UTI) was present in 9 patients (39.1%), 3 of them were symptomatic. None had vesicoureteric reflux (VUR). No anatomical anomalies were detected on imaging studies.

Calculi were located in the left Kidney of 13 patients (56.6%), in right kidney of 5 patients (21.7%) and bilaterally in 4 (17.4%). One patient had vesical and left ureteric stones in addition to both kidneys (4.3%).

Staghorn calculi were seen in 4 patients (17.4%). It was bilateral in 2 of them; with recurrent UTI while it was unilateral in the other 2 with sterile urine. Stones were analyzed from 10 patients (43.5%). They were composed of Cystine in 4, mixed Cystine with Calcium Phosphate or Oxalate in 4, Cystine with Uric acid in 1 and mixed Cystine, Uric acid and Calcium Phosphate in 1, see table 2. In 4 out of 6 patients with mixed calculi there were associated metabolic disorders (66.6%). Overall associated HUr was recorded in 7 patients (30.4%), HCa in 3 (13%) and HOx in one (4.3%).

The length of follow up ranged from 1 to 88 months (median = 9 months and mean = 28.7 ± 6.6 months). No follow up information were available in 3 patients. Ten patients were observed for 3 years.

Nine patients were treated with increased oral fluids and alkali. Three of those 9 patients were stone free at beginning of this therapy. In 1, the stone

passed spontaneously and in 2, stones dissolved as a result of prior DP therapy. All 3 remained stone free with fluid and alkali therapy. Six patients had X–ray evidence of calculi when this therapy was initiated. Two of these patients became stone free. Stone size was decreased in one patient. In 2 patients stone size was not changed and in one patient, the stone was increased in size.

Stone free rate for this therapy was (55.6%), recorded in 5/9 patients.

See table 3. Two patients with unchanged stone size or continued growth were found to have associated HCa + HUr. It was noticed that patients responding to this therapy either as stone free or decreased stone size have a milder disease in term of single stone, small stone (less than 5 mm) and no recurrence. Length of follow up for the patients on fluid and alkali therapy ranged from 7 – 42 months (median = 12 months and mean of 18.4 ± 4.9 months)

Thirteen patients with radiologically proven calculi (two of those 13 had bilateral renal stones) received DP in addition to fluid and alkali. Three of them received DP twice after recurrence of stones. Therefore DP therapy was instituted in 18 occasions. The duration of DP therapy ranged from 2 to 54 months. One patient received DP for 11 years before being involved in this study. Two patients were lost to follow up. In 4 occasions (22.2%), treatments were stopped because patients developed side effects to DP.

Accordingly only 12 occasions of DP treatment on 7 patients were evaluated. In 7 of those 12 occasions (58.3%), stones dissolved. Two out of 12 stones were reduced in size. One stone was unchanged in size and 2 stones continued to grow (Table 2). Length of follow up the patients on DP therapy ranged from 7 – 63 months (median = 31.5 months and mean of 32.5 ± 7.4 months).

Two patients with increasing stone size had associated metabolic disorders.One patient had HUR, the other patient had HUR + HCa. Chemical analysis confirmed mixed Cystine with Calcium Phosphate stone. The patient with unchanged stone size was not regularly complying with medication.

Of the 7 patients evaluated for DP therapy, 3 were continued on DP because of recurrence. In the other 4 patients, DP was stopped after dissolution of their stones.

Recurrence of new stones was detected within 2 and 4 months after stopping DP in 2 of them. Side effects to DP were recorded in 4 patients. Two patients developed thrombocytopenia, one developed nephrotic Syndrome and one patient complained of severe epigastric pain. All symptoms disappeared upon withdrawal of the drug but reappeared upon its recommencement.

Four patients received Captopril. Response was evaluated after 6 months. In 2 patients, stones increased in size. In one patient stone size was unchanged. In one patient stone mass was reduced (Table 2).

Eight patients underwent renal ESWL. Four of them had ESWL on both kidneys. Therefore, 12 ESWL treatments were evaluated.

Stone free rate for renal ESWL was detected in 6 treatments (50%). Reduction in stone size or unchanged stone size was seen in 3 treatments (25%) for each (Table 2).

From 3 patients with unchanged stone size, 2 had concomitant Captopril therapy and one had Staghorn calculus with recurrent UTI. The Highest number

of ESWL treatments was 7 on a left kidney in a patient with recurrent bilateral calculi.

Three ESWL treatments were performed in each of 5 patients. One ESWL treatment was performed in each of 3 patients. Length of follow up of the patients on ESWL ranged from 4 – 80 months (median = 47 months and mean of 40.9 ± 8.7 months)

Twelve patients had a total of 18 open surgical removals of stones. Seven (38.9%) surgical procedures were performed in 5 patients before referral to Al–Kadhymia hospital. One of those 5 had 3 open surgeries on the left Kidney.

Thirteen (72.2%) operations were renal, 3 (16.7%) on the ureter and 2 (11.1%) on the Bladder. Fifteen surgeries were done before medical therapy while 3 were done after medical treatment.

From 20 patient's follow up data, 12 patients (45%) required combined treatments in which more than one treatment modality was used. Fourteen patients developed 27 episodes of recurrence of stone disease (70%), one of them had 4 episodes of recurrence, 4 had 3, 2 had 2 and 7 patients had one recurrence.

Table 1: Clinical presentation of 23 patients with cystinuria

Clinical presentation	*No. of patients*	*(%)*
Passing stones/ gravels	**12**	**52.2**
Gross hematuria	**11**	**47.8**
UTI	**9**	**39.1**
Abdominal Pain	**8**	**34.8**
Dysuria	**6**	**26.1**
Failure to thrive	**4**	**17.4**

*Patient may had more than one clinical presentation

Table 2: Stone chemical analysis of 10 patients

STONE COMPOSITION	No.
Pure Cystine	**4**
Cystine + Calcium Phosphate or Calcium Oxalate	**4**
Cystine + Uric acid	**1**
Cystine + Uric acid + Calcium Phosphate	**1**

Table 3: Treatment modalities and clinical outcome

Treatment	*No. of treatments*	*Stone free*	*Reduced in size*	*No change in size*	*Increased in size*
Fluid and Alkali	**9**	**5 (55.5%)**	**1 (11.1%)**	**2 (22.2%)**	**1 (11.1%)**
D Penicillamine	**12**	**7 (58.3%)**	**2 (16.7%)**	**1 (8.3%)**	**2 (16.7%)**
Captopril	**4**	**0 (0%)**	**1 (25%)**	**1 (25%)**	**2 (50%)**
ESWL	**12**	**6 (50%)**	**3 (25%)**	**3 (25%)**	**0 (0%)**

Discussion

The genetic transport defect in cystinuria exists from birth and stone formation begins in the first decades of life in a rate of 80% and continues life long. [3, 4, 9, 10] This was reflected in our study by early onset of stone disease among the studied group. Some data reported equal incidence between sex [1, 8], other studies reported higher rates among males [5, 17]. Males often have early appearance of stones, more severely affected and produce significantly more stones than females [1, 4, 8], these findings might lead to early presentation and diagnosis of the disease and will be reflected as higher rates in males.

High rate of parental consanguinity which is common in our community and family history of stone disease is obvious because of autosomal recessive inheritance pattern of the disease [1, 3, 4, 6, 7, 8, 12]. Dahlberg et al reported 50% of family history of stone disease among his series [5].

An interesting finding was the presentation of 4 patients with failure to thrive in our study. Andrwes was the first to suggest that cystinuric patients were shorter than normal, Colliss and associates also reported that the average height of 44 cystinuric patients was 2.5 cm less than that of the normal population, he postulated that patients with Cystinuria were at slight nutritional disadvantage as a result of urinary losses and defective gastrointestinal absorption of Lysine, an essential amino acid [5]. Al–Hermi et al described failure to thrive as the presenting feature in 1 of 3 cystinuria cases in Bahrain [18]. A recent epidemiological study from UK revealed that the median height and weight of children with renal stones on presentation was lower than average, claimed that those children often have feeding and growth problems [19]. Concurrent illness may also be contributing factors for reduced growth as we noticed in our study.

Similar to our results, associated UTI was recorded in one third of Cystinuric patients by several repots, as the presenting complaint or as a complication of stone disease [1, 3, 5, 6, 8]

Infection with urease splitting organisms represents a major reason for failure of medical therapy and the need for surgery to eliminate all fragments of the calculus in order to sterilize the urinary tract [6].

Staghorn calculi are well recognized radiological appearance with Cystinuria [1, 9, 10, 18]. The permanent excretion of excessive amount of Cystine is spontaneously associated with the relentless formation of stones which can have a staghorn development [3]. Most often, these stones now are treated with combination of percutaneous nephrolithotomy / ESWL / second look nephroscopy, termed "sandwich therapy " ,which precludes the need for renal open surgery [1, 3, 4, 18]. Nearly similar to our results, pure cystine stones were observed in 60 – 80% of cases [1, 4].

Sakhaee et al observed that HCa, HUr and/ or Hypocitraturia may accompany Cystinuria, thus contributing to stone formation [3] Avoidance of foods with a very high Methionine content; which is the precursor of Cystine, could lower urinary Cystine excretion, achieving one goal of treatment. However; most authors believe that such dietary restriction is not advisable for children [3, 4, 12]. Low salt diet to about 1 mmol / Kg / day was effective in reducing urinary Cystine concentration in children, however long term

compliance even with modest salt restriction is often poor[3, 4, 12]. Because high sodium intake increases cystine excretion, potassium citrate was preferred for most of the studied patients [1, 3, 8, 10].

A high fluid intake remains the most important factor for the reduction of urinary Cystine concentration which should be distributed throughout day and night [3, 4, 7, 10]. It was calculated that urine excretion of > 3 – 4 Liters/ day (1.2 – 2 liters/ M^2/ day or 40 ml/ Kg/ day in children) is necessary to prevent stone formation [1, 3, 9, 12]. Urine dilution alone is rarely sufficient and sustained alkalinization to PH of about 7.5 is often required to obtain effective Cystine solubilization [3, 4, 7, 10, 12]. The first stone dissolution by alkilinization of urine was reported in 1924, and alkilinization has been the mainstream treatment for over 50 years [9]. Alkaline urine can prevent precipitation of cystine calculi and even aid in dissolution [1]. Dent et al assessed the efficacy of large urine volume with alkali (Sodium or Potassium Bicarbonate) on Cystinurics. He reported that 12 of 18 stones of his series either dissolved or patients remained stone free [9]

In a study from Japan, in 3 cases out of 15 children with cystinuria, calculi disappeared by medical therapy alone [17].

We could not correlate our results with urine Cystine level; due to lack of laboratory resources. In Mayo clinic series; successful therapy was obtained in 10 out of 30 patients treated with oral fluid and alkali only [5].

For stone free patients; first line therapy is the conservative approach, is adequate for stone prevention [1, 3, 5]. This observation was highlighted in our study as 3 patients who did not have stones at onset of conservative therapy, continued to be stone free.

When conservative measures fail, the application of chelating agents that transform Cystine into a highly soluble Thiol–Cysteine mixed disulfide may be added. The most widely used drugs are DP and α – mercaptopropionyl – glycine or Tiopronin (TP), both drugs are equally effective [3, 4, 7 – 10, 12].

DP was introduced as a treatment for Cystinuria in 1963 [9]. Over the next few years, there were many small published series demonstrating reduction in Cystine excretion, stone dissolution and reduction in recurrence not controlled by oral fluids and alkalinization [9]. DP is best suited for stone prevention after surgical debulking of stone burden [1]. This observation was noticed in 4 occasions in our study. The same was observed in 3 out of 24 patients in Mayo clinic, where 10 patients had complete dissolution of calculi. Stone mass was reduced in 5, no change in stone size in 3 and stones continue to grow in another three [5]. Prevalence rate of adverse reactions to DP is approximately 20 – 50% in several studies [1, 3, 4, 5, 7 - 10]. Higher rate of side effect (85.7%) to DP was reported in a Japanese Study [17]. Coexisting metabolic disorders and presence of mixed calculi were emphasized as the most common reason for failure of DP therapy [3, 5] TP is preferred by most physicians in treatment of cystinuria, because of it's less side effects and can be used in patients who develop allergic reactions to DP [1, 3, 4, 8 – 10] Being unavailable in our country, TP was not used for the studied group.

Compared to the small series in this study, several studies failed to observe a significant effect of Captopril on Cystine stones formation [3, 9, 20].

Because of its low morbidity, ESWL can be offered as first line treatment for Cystine stones in children [1, 4].

Cystine stones often require urological intervention when they cause symptoms, obstruction, and increase in size [1, 3]. Although Cystine stones respond less to ESWL than Calcium stones, the stone free rate after ESWL is higher even for large ones in children than adults[4]. Because of their hardness and homogenous amino acid composition, most Cystine stones require multiple cessions to achieve acceptable stone free rates [1, 3]. This observation was also highlighted in this study. The response to ESWL is recorded in this study as the final result of completing treatments. Medical treatment was used in all patients underwent ESWL.

Stone free rate following ESWL was 43% in a Japanese study with an average of 5.9 sessions [17].

Open surgery should be considered for cases with complex staghorn calculi, difficult anatomy and simultaneous urological abnormalities [4]. As many patients will suffer recurrence of stones within a few years and need repeated urological intervention, minimally invasive procedures should be preferred to open surgery whenever possible [1, 3] . In a study from Japan, 17 lithotomies were performed in 13 out of 15 patients [17]

Treatment of patients with Cystinuria requires close co operation between the urologist and the nephrologists [1, 3, 4].

Regular medical treatment is mandatory because of the relentless tendency of cystine stones to recur despite improved urological techniques of stone removal [1, 3].

The main deficiency of this study is in the inability to quantitatively measure urine cystine, an important tool in the assessment of treatment efficiency. However it was believed that fellow up on stone formation rate provide a real picture of the clinical outcome.

In conclusion, oral fluids and alkali were most successful when used prophylactically in the stone free patients. D-Penicillamine and ESWL had nearly equal rate of successful results. Failure of treatment should alarm the possibility of coexisting metabolic disorders and UTI as a complicating problem. Combined treatments were frequently required in cystinuric children.

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Cd30 molecule expression in sera and on t cells of trophoblast tissue from women with recurrent spontanious abortion

Nidhal Abdul-Mohymen[1] *PhD*, **Amal Hussain[2]** *PhD*.

Abstract

Background: Immune responses during pregnancy show a distinct shift towards Th2-type reactions occurs, especially at the feto-maternal interface. CD30 has been described as being preferentially expressed, and released, by human T cells producing the Th2-type cytokines**.**

Objective: To determine the level of soluble CD30 (sCD30) in serum and in the trophoblasts of patients with recurrent spontaneous abortion (RSA).

Materials and methods*:* A total of sixty one women attending the Obstetrics department in al- kadhemia hospital, age range from (23.9 - 28.5years), were enrolled in the current study and were further classified into three categories: Group A: 35 women included cases with recurrent spontaneous abortion, group B: 16 women included non- recurrent spontaneous abortion (non-RSA): group C: 10 women was Control (successful pregnancy). From each subject blood sample and placental tissues were collected and serum was seperated for the estimation of soluble CD30 (sCD 30) levels using ELISA method and trophoblasts tissues (an image for the local microenvironment) were screened to determine the levels of CD30 using immunohistochemistry.

Results: Trophoblast expression of CD30 and sCD30, showed a highly significant increased values for both patients groups ($p<0.001$) when compared with control group.

Conclusion: It is likely that there may be an association between normal pregnancy and CD30 density on the cell surface.

Key words: recurrent spontaneous abortion, CD30, mmunohistochemistry, ELISA

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Introduction

Successful human pregnancy appears to be an immunological paradox, in that the fetus represents a semi-allograft developing in the potentially hostile environment of the maternal immune system [1-3] One important mechanism involves the down-regulation of the cellular immune response, which has been shown to be dependent upon the suppression of T-helper (Th)1 and T-cytotoxic (Tc)1cells,which produce interleukin(IL-2), interferon (IFN)-γ, and tumor necrosis factor (TNF)-β, and the up-regulation of Th2 and Tc2 cells, which produce IL-4, IL-6, IL-10 and IL-13[4-7].

Previous investigations of Th1/Th2 immune responses during pregnancy were able to show that a distinct shift towards Th2-type reactions occurs, especially at the feto-maternal interface [8-12]. CD30 has been described as being preferentially expressed, and released, by human T cells producing the Th2-type cytokines [13,14]. Surface CD30 is cleaved proteolytically, resulting in the release of the soluble form of the molecule (sCD30) by CD30-expressing cells [15].

Since CD30 has been reported to be associated with Th2-type reactivity, we measured soluble CD30 in the serum and the density of CD30 on the surface of T cells of normal pregnant women and in women undergoing abortion ; to clarify if there is any association between normal pregnancy

[1]Dept. Medical Microbiology, College of Medicine, Al-Nahrain University, [2] Dept. Medical Microbiology, College of Medicine, Al- Mustansiryia University.

Adress Correspondence to: Dr. Nidhal AbdulMohymen.

E- mail: dr.nidhalmohammed@yahoo.com

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and CD30 density on the cell surface we designed this study.

Subjects, materials and methods

Sixty one women attending the Obstetrics and Gynecology department of Al-Kadhemia Teaching Hospital in Baghdad between December 2004 and August 2005 were the subject of studying. They included 35 women with 3-6 consecutive aborti**n; group A** (RSA), 16 women with abortion for the first or second time; group B, 10 pregnant women who had at least two previous normal pregnancies;group C.

Trophoblastic tissue was collected from the evacuation of retained pieces during the procedure of curettage and placed in 10% formaldehyde. Two to three paraffin embedded blocks were prepared for each patient. Staining with haematoxyline and eosin was carried out to decide which block can be used in the study (only sections that contained trophoplastic tissues were included. These samples were subjected for immunohistochemical staining (IHC) protocols with the anti CD30 marker according to [16]. The expression of CD30 was measured by counting the number of positive trophoblastic cells that gave brown cytoplasmic staining under light microscope. The extent of the IHC signal in the villi was determined in 10 fields (100 X magnification). In each field the total number of villi were counted and the extent of cytoplasmic staining of the trophoblast cells in a given villous was determined as a percent. The total staining score was divided by the number of whole villi per field in 10 fields [17] so, the percentage of positively stained villi in the 10 fields was calculated for each case by taking the mean of the percentage of the positivity stained villi in the 10 fields. In each field the total number of villi was counted and the extent of staining of the cytotrophoblast and syncytiotrophoblast in a given villous was scored as: score 3 (75-100%); 2 (25-75%); or 1 (<25%).

Detection of soluble CD30 by ELISA:

Sample collection: Five ml of venous blood was collected from each subject group and serum was separated and stored at -20 ^{o}C until used.

The ELISA test was perfomed,using two anti- CD30 monoclonal antibobies (Primary and secondary)antibodies which were the product of DAKO.Cut –off value was calculated according to[18].

Statistical Analysis

The ANOVA analysis program, chi-square and the relationship between the indicators was measured qualitatively by using the correlation coefficient.

Results

Correlation between abortion and CD30:

In thirty five women with RSA (group A), a negative significant correlation (p<0.05) between abortion and CD30 in sera and trophoblast tissues (r=-0.651; r=-0.496, respectively), was found, (Table **1**). The data also showed a negative significant correlation(r=-0.529; $p\leq0.05$) between abortion and sCD30 in group (B).

Table 1: Correlation between abortion and CD30 in women involved in this study.

Variables	Groups	n	Correlation Coefficient r =	*P* value≤
Abortion– sCD30(ELISA)	**A**	**35**	**-0.651**	**0.05**
	B	**16**	**-0.529**	**0.05**
	C	**10**	0.510	N.S.
Abortion– CD30(IHC)	**A**	**35**	**-0.496**	**0.05**
	B	**16**	0.297	N.S.
	C	**10**	0.430	N.S.

N.S. =not significant

Table 2: Number and percentage of CD30 in trophoblasts of studied groups.

Variable	Score*	Groups			Chi-Square *P* value
		A	B	C	
		n=35 (%)	n=16 (%)	n=10 (%)	
CD30 (IHC)	1	16(45.7)	1(6.3)	0	0.001**
	2	19(54.3)	15(93.7)	5(50)	
	3	0	0	5(50)	

*In each field the total number of villi were counted and the extent of staining of the cytotrophoblast and syncytiotro-phoblast in a given villous was graded as 3 (75-100%); 2 (25-75%); or 1 (<25%), **= highly significant difference ($p<0.01$)

Expression of CD30 in villous trophoblasts detected by IHC:

The results showed that percentage of CD30 expression was moderate in 54.3% (19/35) and 93.7 %(15/16) of women in group A and group B, respectively.The corresponding figure in control group was 50%. These differences were highly significant (P≤ 0.001), (Table 2).

In (Table 3), The mean percentage of CD30 expression in the trophoplast was significantly declined ($p<0.001$) in group A as compared with group C (23. 7± 1.1vs 76 ± 3.3) respectively.The decline was also found in group B as compared with group C (39.6± 2.5 vs 76 ± 3.3) respectively.

Table 3 : Comparison between the mean percentages of CD30 expression in the trophoblasts of the studied groups.

Variable	Group	No=61	Mean± SE	F test *P* value	P values between groups	
					Groups	*P* ≤
CD30 (IHC)	A	35	23.7.±1.1	<0.01	A–B	0.001
	B	16	39.6±2.5		A –C	0.001
	C	10	76± 3.3		B –C	0.001

Soluble CD30 (sCD30) in sera detected by ELISA:

Table 4 shows that the mean value of serum levels of sCD30 was significantly higher (p<0.001) in group A (0.125±0.01) and B (0.127±0.01) as compared with group C (0.61± 0.06).

Table 4: Comparison between the mean values of sCD30 in sera of women involved in the study.

Variable	Group	No.=61	Mean±SE	F test *P* value	Pvalues between groups	
					groups	*P* ≤
CD30 (ELISA)	A	35	0.125±0.01	<0.01	A–B	NS.
	B	16	0.127±0.01		A –C	0.001
	C	10	0.61± 0.06		B –C	0.001

NS. =not significant

Discussion

The current study, showed highly significant increase in expression of CD30 (local and systemic) (p<0.001) in control group (successful pregnancy) compared with first and second trimester abortion. In addition, a significant difference in expression of CD30 (p<0.05) between first trimester and second trimester abortion was found. These results might be explained by the presence of association between normal pregnancy and CD30 density on the cell surface in trophoblast and circulation.

Results of group A, found a negative significant correlation between gestational age and CD30 in circulation and trophoblasts tissue(r=-.651; r=-.496, p<0.05, respectively), and gave a negative significant correlation (r=-.529; p<0.05) between gestational age and sCD30 in women in group B. This result indicated that in women with RSA the expression of CD30 decreasing with increasing gestational age. This might be explained by other studies showing that high concentrations of sCD30 have been found in a variety of disorders that are clearly Th2-mediated or Th2-dominated [13]. In addition, the current study, showed highly significant

difference (p<0.001) between CD30 expression in trophoblasts tissue in three groups of investigated women. However, previous reports showed that CD30 expression was associated with differentiation and activation pathway of human T cells producing Th2 –type cytokine [14, 19].

Previous reports showed that CD30, was a member of tumor necrosis factor receptor superfamily TNFR [19] and can give signals through the activation of the nuclear factor-κB (NFκB), which is an important transcriptional factor, regulating the pro-inflammatory cytokines, which were shown to be down-regulating during pregnancy, a mechanism that is essential for the maintenance of the Type2 cytokine profile required for pregnancy success [20]. High concentrations of sCD30 have been found in a variety of disorders that are clearly Th2-mediated or Th2-dominated. Since pregnancy appears to be a Th2-biased condition it is likely that the skew towards Th2-bias seen in peripheral blood cells may be reflected by increased concentrations of sCD30 in the blood [13, 19].

Moreover, the expression of CD30 in trophoblatic tissue was highly significant increased (p<0.001) in women with non-RSA (group B) compared with RSA (group A), but no significant difference (p>0.05) was found in the levels of sCD30between the mentioned groups. This result might be associated with level of cytokine and CD30 within local microenviro-nment and the peripheral circulation. The variation of expression suggests a possible role for hormones, preferably progesterone, in the regulation of CD30 expression , This would be a novel mechanism of CD30 induction [21] , progesterone produce an immunomodulatory protein known as progesterone –induced blocking factor(PIBF) which induces increased production of Th2 cytokines [22], therefore apart from the systemic changes in the maternal immune response, local immunomodulation at the feto-maternal interface via wide array of hormones and cytokines, and immune effector cells also play a critical role in maintaining the balance of the desirable immune response [23]

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Pattern of mycobacterium tuberculosis drug resistance in previously treated cases in Iraq

Mustafa Nema *FICMS; CABMS* ,**Hashim M. Al-Kadimy** *FRCP*.

Abstract

Background: The resistance of certain *Mycobacterium tuberculosis* strains to anti-tuberculosis drugs is not a new phenomenon. It is man-made amplification of natural phenomenon.

Objectives: 1- To provide scientifically based information on the burden of Mycobacterium tuberculosis drug resistance in Iraq. 2- To compare the pattern of this resistance in Iraq with that in the other countries.

Methods: A total 411 patients with pulmonary tuberculosis who have already received at least one month of anti-tuberculosis therapy were selected. Sputum cultures and drug sensitivity tests for Mycobacterium tuberculosis were arranged.

Results: Resistance to rifampicin, isoniazid, streptomycin and ethambutol was noted in 52 (24.4%), 22 (10.3%), 21 (9.9%) and 8 (3.8%) of isolates respectively. Multidrug and four-drug resistance was found in 52 (24.4%) and 24 (11.3%) respectively. Rifampicin resistance in any form was noted in 146 (68.5%).

Conclusion: The magnitude of *Mycobacterium tuberculosis* drug resistance in Iraq found to be relatively high.

Key wards: tuberculosis; antituberculous drugs resistance; multidrug resistance; drug resistance

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Introduction

The emergence of resistance to antimicrobials is a natural biological occurrence. Drug resistance can be divided to: **Drug resistance among new cases (primary resistance)** referred to presence of resistant isolates of tuberculosis (TB) in patients who deny or having no evidence of, prior anti-TB treatment (for as much as 1 month), **drug resistance among previously treated cases (acquired resistance)** which is the presence of resistant isolates of tuberculosis in patients who, in response to direct questioning, admit having been treated for tuberculosis for period of 1 month or more, and **combined prevalence of drug resistance** which is the prevalence of resistance in the population surveyed regardless of prior treatment[1]. *Polyresistance* means resistance to more than one drug other than multidrug resistance.

In 1995, WHO estimated that 50 million people were infected with drug-resistant strains of *Mycobacterium tuberculosis* [2]. In 1997, the World Health Organization (WHO), the International Union against Tuberculosis and Lung disease (IUATLD) and several partners worldwide release the first report of Global Project on Anti-tuberculosis Drug Resistance Surveillance (DRS). This report presented data from 35 geographical settings (surveyed between 1994 and 1996) using standard epidemiological and laboratory guidelines. These data covered 16% of the world notified TB cases.

Drug resistance is less likely to occur in directly observed therapy (DOT) [3]. The survey in Islamic

Al-Kadimia University Hospital.
Adress Correspondence to: Dr. Mustafa Nema.
E- mail: mustafanema71@yahoo.com
mobile: +964 7707 882 782
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Republic of Iran demonstrated high prevalence of multidrug resistant-TB (5%) and any rifampicin (R) resistance (50%). Drug resistance is not a problem in Oman, nor is it in Morocco. Iraq was not included in this project.

The majority of isoniazid (H) resistant strains show mutations of the *katG* gene (50%) followed by mutation locus resistance in *inh*A (25%) of total H resistance. Mutation loci for rifampicin resistance is rpoB (97%), for streptomycin (S) is rpsL (60%), for ethambutol (E) is embAB (50%) and for pyrizinamide (Z) is pcrA with unknown revalence[4,5].

Methods

A total of 411 randomly selected cluster of patients included. All have pulmonary TB and had already received at least one month of anti-TB therapy. The study was conducted from February 2005 to August 2006. All patients selected were sputum positive isolates of *M. tuberculosis* (using standard Zeihl-Nelson stain method). Culture and sensitivity then arranged for patient's sputum samples.

Setting: Institute of tuberculosis and chest disease in Baghdad, receiving patients from all hospitals all over the country, and the first treating facility for a large population in Baghdad. It is the only facility that has performs culture and drug susceptibility testing for *Mycobacterium tuberculosis* (MTB) in Iraq.

Bacteriological methods

Löwenstein-Jensein (L-J) culture medium was used for culturing sputum. Identification of the strains was based on the niacin production test, the nitrate reduction test, the amniobenzoic acid (500mg/l) and the thiophene carboxylic acid (2mg/l) resistance test. Species other than the pathogenic species of MTB complex were excluded from analysis, e.g. rapid grower species that grow within three days.

Cultures that yield MTB then subjected to drug sensitivity tests. Drug sensitivity test was performed using the proportion method on L-J medium.

Using the proportion method, resistance was expressed as percentage of colonies that grew on critical concentrations of the drugs which were prepared as follows:

H: 0.2 µg/ml, R: 40 µg/ml, E: 2 µg/ml and S: 4 µg/ml. The criterion for resistance to a particular drug was growth of 1% of the population on medium containing the critical concentration. The results of the tests recorded on standardized laboratory forms.

Radiometric BACTEC MGIT 960 (Becton Dickinson) method, started to be in work since 2002 in Iraq, used for both mycobacterial detection and drug susceptibility testing. Using this method, resistance was expressed as growth concentration in each tube containing the following concentrations (compared to growth control): H: 0.1 µg/ml, R: 1 µg/ml, E: 5 µg/ml and S: 1 µg/ml.

Screening for the human immune deficiency virus (HIV) was not part of this study.

Statistical analysis

SPSS ® version 10.0.5 for Windows to analyze the data. The Student t-test was used to calculate continuous variables. A *P* value of 0.05 was considered significant.

Results

Three hundred-eleven (73%) were males and 100 (24.3%) were females. The median age of the patients was 34 years. Mean age = 35.4 ± 10.4 years with a male: female ratio of 3:1. The age group most commonly encountered in

this study range between 25 to 44 years: 206 (50%) males and 70 (17%) females. The median age of patients **with resistant** MTB was 34 years (range 20-65 years) for males and for females was 35 (age range 20-54 years), while for those **with sensitive** isolates the median age was 34 years for males(age range 20-67 years), and for females it was 35.5 years (age range 21-68 years) (Table 1).

One hundred eighty-nine (48.2%) isolates were sensitive to H, R, S and E, and 213 (51.8%) were resistant to at least one of the four first-line agents. Multidrug resistance was found in 52 (12.6%) isolates. Four-drug resistance was highest among the MDR group, noted in 24 (5.8%). Any drug resistance noted in 213 (51.8%) (Table 2) (Figure 1and 2).

Monoresistance was noted in 103 (25.1%). As monoresitance, resistance to R was the highest, noted in 52 (12.6%) isolates; H resistance was the second most common, found in 22 (5.3%), S resistance was noted in 21 (1.9%) and E resistance was the lowest, in 8 (1.9%) isolates. Combined resistance of S with R and in other hand S with H was found to be higher than other types of polyresistance, 20 (4.9%) and 10 (2.4%) respectively. Rifampicin resistance, in any form, exceed any other type of resistance where it was noted in 146 (35.5%), followed by H in 103 (25.1%), S in 93 patients (22.8%) and E in 47 patients (11.4%) (Table 2) (Figure 2).

When compared to other geographical settings, drug resistance in Iraq outweigh that in Saudi Arabia in (a) *any resistance*: (51.8% compared to 22%), (b) *1, 2 and 3 drug(s) resistance*: (26%, 14% and 7% compared to 6%, 6% and 2%) and (c) *MDR*: (12.6% compared to 2%). While four drug resistance in Iraq is lower than that in Saudi Arabia: 6% compared to 8%.

In comparison to Islamic Republic of Iran, overall resistance in Iraq was *lower*, (51.8% compared to 57.1%), as well as 4-drugs: (11.3% compared to 28.6%) and MDR (12.6% compared to 48.2%) respectively, while resistance to 1, 2, and 3 drugs were *higher*, (48.45, 25.4%, 12.2% compared to 7.1%, 14.3%, 7.2%) respectively (Table 3) (Figure 3).

Table 1: Characteristics of the study sample

Sensitivity	Gender	Mean age	Median age	Age range
Sensitive	Male	34.9 $\pm$ 10.5	34	20 – 67
	Female	37.2 $\pm$ 10.9	35.5	21 – 68
Resistant	Male	35.3 $\pm$ 10.6	34	20 – 65
	Female	35.5 + 9.5	35	20 – 54

Table 2: Patterns of anti-tuberculosis drug resistance (in previously treated cases)

	Number	%
Total tested	411	100
Fully sensitive	198	48.2
Any resistance	213	51.8
Mono-resistance	103	25.1
H*	22	5.3
R**	52	12.6
E***	8	1.9
S****	21	5.1
H + R resistance	52	12.6
HR	15	3.6
HRE	9	2.2
HRS	14	3.4
HRSE	24	5.8
H + other resistance	12	2.9
HE	2	0.5
HS	10	2.4
HES	0	0.0
R + other resistance	28	6.8
RS	20	4.9
RE	5	1.2
RES	3	0.7
Other multi-resistance	2	0.5
ES	2	0.5
Any H resistance	103	25.1
Any R resistance	146	35.5

*H= isoniazid, **R= rifampicin, ***E= ethambutol, ****S= sreptomycin

Table 3: Pattern of drug resistance to each drug among previously treated cases in three geographical settings (figures in percentage)

Country	Year	No. of patients tested	Overall		Resistance to:				Poly-resistance	
			Susceptible %	Resistant %	1Drug %	2Drugs %	3Drugs %	4Drugs %	Any %	Multidrug resistance %
Iraq	2006	411	48.2	51.8	48.4	25.4	12.2	11.3	14.2	12.6
Iran	1998	666	42.9	57.1	7.1	14.3	7.2	28.6	50	48.2
Saudi Arabia	2004	764	91.5	22	6	6	2	8	2	2

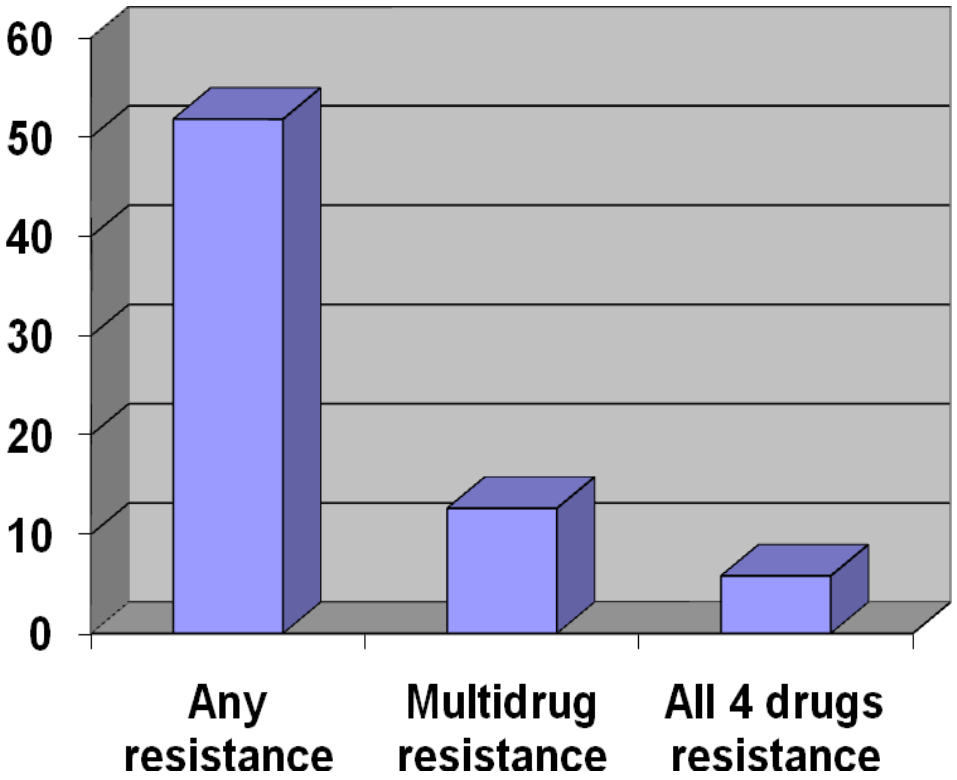

Figure 1: Drug resistance indicators among previously treated cases

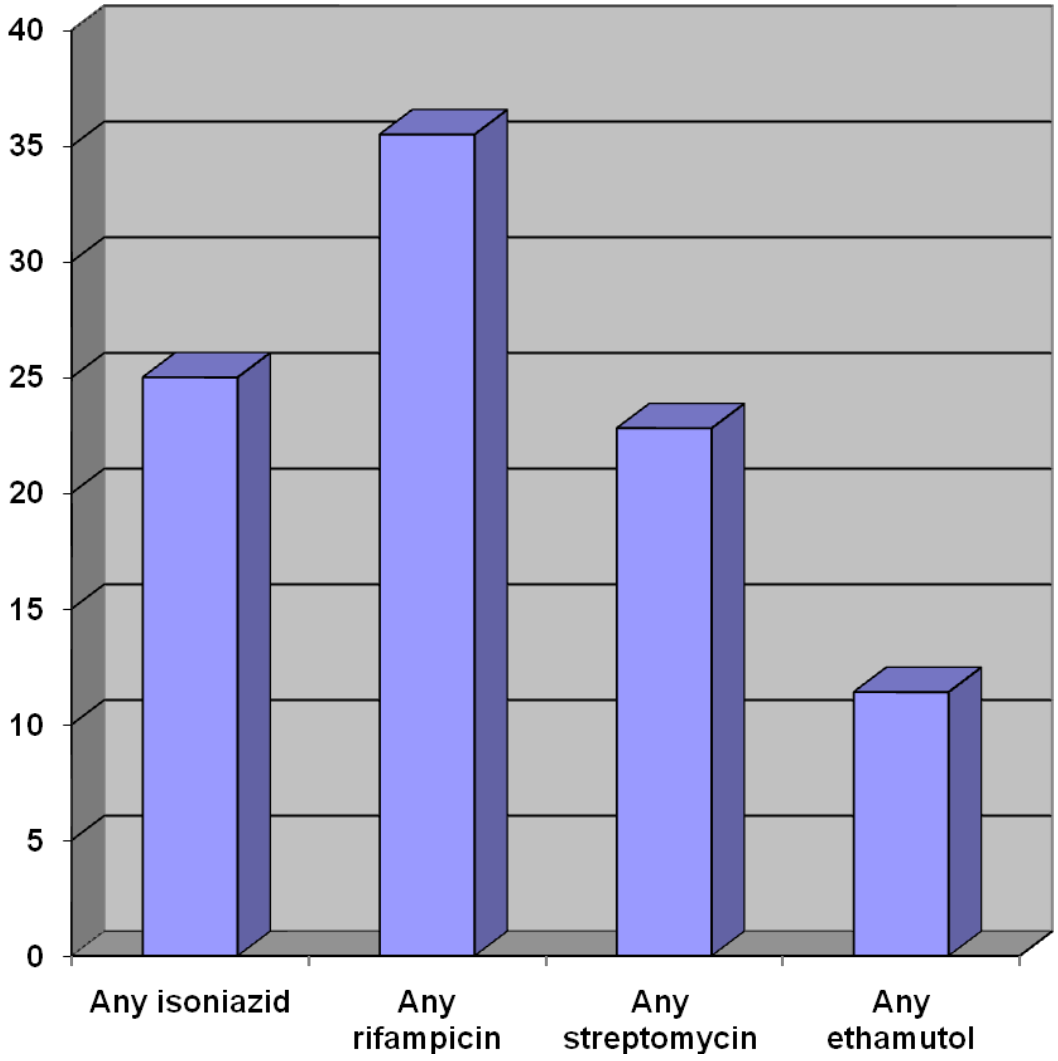

Figure 2: Prevalence of any drug resistance among previously treated cases according to specific drugs

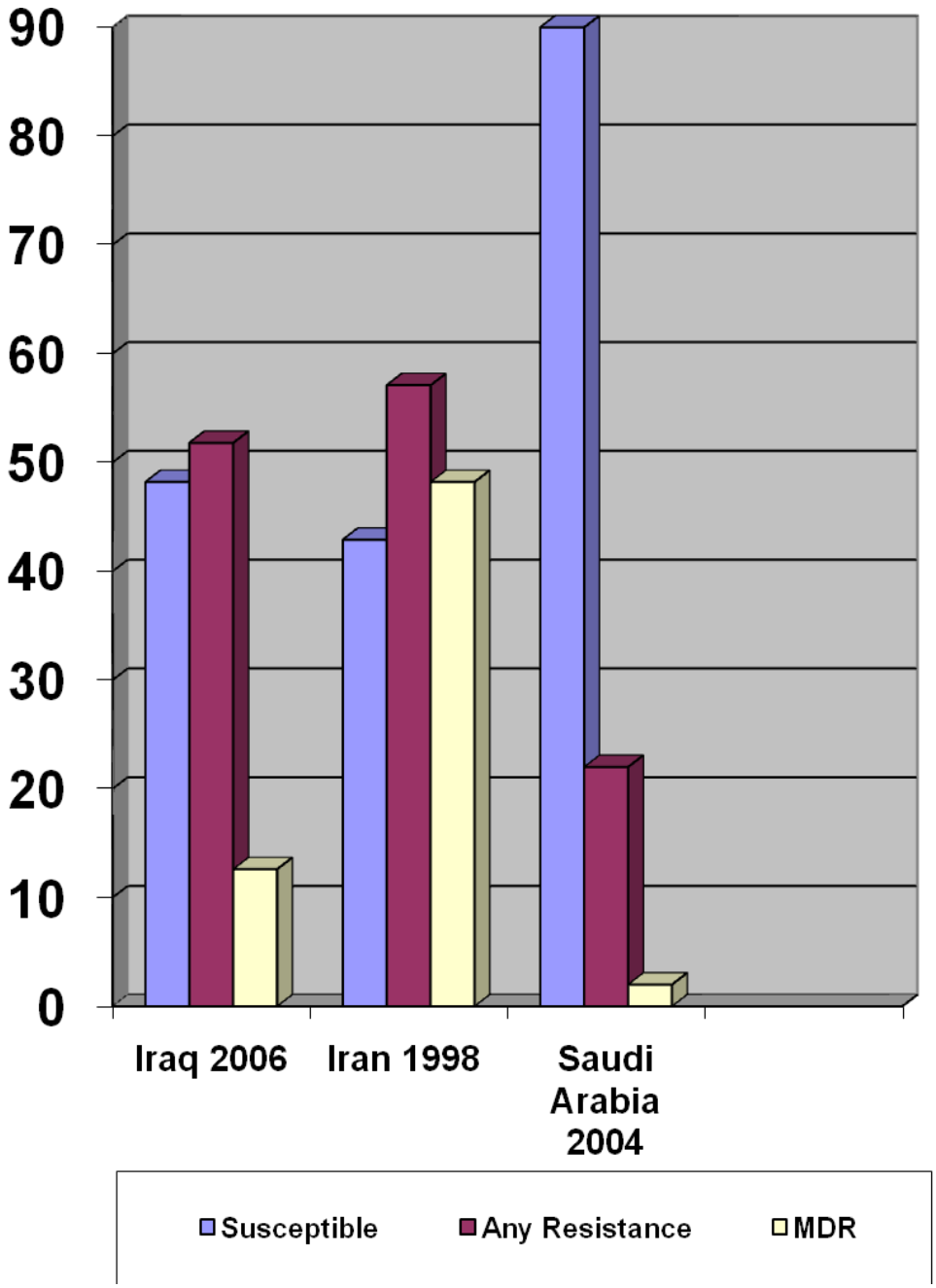

Figure 3: Pattern of multidrug resistance, any drug resistance and susceptible cases in three geographical settings

Discussion

Reports of drug resistance of *M. tuberculosis* in Iraq are limited. The prevalence of drug resistance to one drug (**monoresistance**) was 25.1%, while that of resistance to **all four drugs** was 58%. Monoresistance is higher and four-drug resistance is lower than that in Islamic republic of Iran, and both are higher than that in Saudi Arabia. In the Global Project the median prevalence of drug resistance to one drug was 11.3, and the median prevalence of resistance to all

four drugs was 1.8 [6].

Any drug resistance among previously treated cases found to be 51.8% which is higher than that in Saudi Arabia (22%) [7] and lower than that in Islamic Republic of Iran (57.1%). This type of resistance falls in the median value when compared to other countries in Global Project which is ranged from 0% in Finland to 93.8% in Uruguay with a median prevalence of 23.3 %[8].

Increases in prevalence of *any resistance* may reflect an environment that favors the acquisition of additional resistance and can lead to future increases in MDR [1].

High number of drug-resistance among previously treated cases seen in this study may be related to high number of resistant strains circulating the in the community due to very high pool of previously treated cases.

MDR-TB among previously treated cases was 12.6%. It is ranged from 0% in 4 geographical settings in Global Project, to 48.2% in Islamic republic of Iran. The median prevalence in Global Project was 9.3%[9]. In Saudi Arabia it is 14 %[7]. The most powerful predictor of MDR-TB was a history of past treatment, which was usually accompanied by inadequate drug regimen [10]

Monoresistance to rifampicin (R) was (12.6%) outweighing other types of monoresistance (H, E, and S), but is lower than that found Egypt (21.7%)[1] and it was not recorded in Islamic Republic of Iran nor in Saudi Arabia [7,8]. In addition, relatively high prevalence of **any rifampicin resistance** (35.5%), the most powerful anti-tuberculosis drug and a key determinant of MDR-TB, was reported. It is higher than that recorded in Egypt (7%)[1] and Saudi Arabia (14%)[7] but lower than that in Islamic Republic of Iran (50%)[8]

Rifampicin might be used in Iraq, unexpectedly, to treat other types of infections, like brucellosis and geneto-urinary infections as well as incorrectly prescribed or taken by patients - singly or inappropriately combined - with other anti-TB; therefore, high level of resistance to this very important anti-TB drug could be produced.

Any resistance to streptomycin (S) was observed in 22.8%, lower than the value found in Egypt (23.6%)[1] and Iran (39.3%)[8] but higher than Saudi (12%)[7].

Any resistance to ethambutol (E) was noted in 11.4%, which is more than Saudi 10%[7], but lower than Iran 32.1%[8], and Egypt (30.9%)[1].

Polyresistance was noted in 23% of isolates, which is higher than that found in Saudi (1%) [7], and lower than that found in Iran (50%) [8].

When compared to other geographical settings, drug resistance in Iraq outweighs that in Saudi regarding *any resistance* (51.8 compared to 22%), while it is lower than that in Iran (57.1%). Resistance to *one drug* was 25.1 which is more than that in Iran and Saudi (7.1 and 6 respectively). *Tow and three drug* resistance were found to be 13.1 and 6.3%. Both percents are higher than that found Saudi (6 and 2%) and lower than Iran (6% and 14.3 respectively).

In comparison to Islamic Republic of Iran, *overall resistance* in Iraq was lower (51.8% compared to 57.1%), as well as *4-drug resistance* (5.8% compared to 28.6%) and *MDR* (12.6% compared to 48.2%) while resistance *to 1, 2, 3 drug(s)* were higher, (48.45, 25.4%, 12.2% compared to 7.1%, 14.3%, 7.1%), (Table 3) (Figure 3).

From total cases sampled, cases

with *any resistance* exceed those who were sensitive by about 3%. This may be regarded as a **bias** related to the drawback of this study in the way of collecting samples, where patient with TB suspected of having some problem (resistance or other complications), were referred to the single institute from different country cities, therefore, the high number of resistance cases found.

Many factors acting locally, affect TB control and drug resistance prevalence. These include non-use of DOT regimen in Iraq, effect of war and other social and economic limitations.

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Immunophenotyphing of Peripheral Blood Lymphocytes to person Exposed to electromagnetic fields

Rafid Abdul -Wahid *MSc*.

Abstract

Background: There is considerabl evidence relating electromagnetic fields (EMFs) exposure to reduce immune system competence .and these changes associated with cell growth control, differentiation and proliferation of cells of immune system, trans membrane signaling cascades, gap junction communication, immune system action.

Objective: to investigate the Phenotyping of peripheral blood lymphocytes of volunteer's exposed at least 10 years to electromagnetic fields (EMFs) induced by transmission power lines in their residential area ,

Subjects and methods: fourty five volunteer's aged between 25 and 65 Years, exposed for at least 10 years to electromagnetic fields (EMFs) induced by transmission power lines in their residential area and Fifteen male of similar age unexposed, away from the transmission power lines as a control group were used in this study . The electromagnetic fields (EMFs) (with range of 50 Hz) beside the homes of the volunteer's. This study carried out in three different are as of Baghdad included (Al –Bladyat, Hay al-adel and Al-Dorra cities) , The groups of this study were divided into three sub- groups according to the distance away from the towers of transmission power lines (1) range: from 1 to 25 meter (2) from 25 to 50 meter (3) from 50 to 75 meter . Phenotyping of peripheral blood lymphocytes had been done by direct immunofluorescent microscopy using anti –CD 3(for T-cells detection), anti-CD4 (for T- helper-cells), anti CD8 (for T-cytotoxic/suppressor cells), anti CD21 (for B-cells) and anti CD56 (for natural killer cells).

Results: A statistically significant reduction of PBL percentage bearing CD3,CD4,CD19,CD56($P<0.01$) between the exposed volunteers and control ;.except CD8 which showed no significant different between these groups . the mean percentage of $CD4^+/CD8^+$ ratio in exposed volunteers groups was significantly ($P<0.01$) lowered in comparison to control group .

Conclusion: we postulated that the chronic exposure to electromagnetic fields from power lines caused suppression in immune system.

Keywords: Electromagnetic felid, CD4, CD8, CD21, CD56, Lymphocytes, phenotyping

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Introduction

Biology has preceded electronic physics because brains and cells use oscillating ion currents for controlling the release of neurotransmitters and in the cell to cell communication systems. Biological systems detect and respond to external ELF signals using their built-in receiving and decoding systems (cell-to-cell communication) [(1)].

Dept. Biochemical engineering, Al-Khwarizmi college, Baghdad University.
Adress Correspondence to: Rafid Abdul -Wahid
E- mail: rafid_sigma@yahoo.com
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In the last five years a large number of experiments have clearly shown various biological and medical effects of (EMFs) at the cellular level, Both human and animal studies report large immunological changes after exposure to environmental levels of electromagnetic fields (EMFs) [(2)].

There is considerable evidence relating EMFs exposure to reduce immune system competence. Many of these evidence show that EMFs -caused changes in processes associated with cell growth control, differentiation and proliferation of cells of immune system, trans membrane signaling cascades, gap

junction communication, immune system action, rates of cell transformation; which are biological processes of considerable interest to scientists who study the molecular and cellular basis of immune system[3].

Lymphocytes phenotyping is part from the mirror image of the immunity, and can give an idea of the immunological status.

Specific reports from studies on exposures to various types of modern EMFs was found over-reaction morphological alterations of immune cells; epically mast cells, enlarged and profound increased of mast cells in the upper skin profound increases in mast cells in the upper skin layer accompany with degranulation of mast cells (electrohypersensitivity) , presence of biological markers for inflammation that are sensitive to EMF exposure at non-thermal levels; changes in lymphocyte viability decreased the number of NK cells(CD56) and T lymphocytes(CD3) [4]

Andrew and his collages demonstrated that a 6-weeks exposure to EMFs induces a significant decrease of CD3+, CD4+, CD8+, CD19 and Natural killer cells Population Linear regression Analysis demonstrated a dose-response relationship between the changes in the immune functions and the EMFs intensities [5].

Another study showed a significant decline in the absolute numbers and ratios of CD4+/CD8+ lymphocytes in favor of CD8+ cells of cows at farm A housed under a 320 kV transmission line exposed to 3.28 T magnetic fields compared to cows at a distant 198 meter Farm B considered zero exposed [6].

Another study conducted on workers of TV re-transmission and satellite communication center found decrease in the level of serum IgG and IgA; decreased the count of peripheral blood CD8, CD4, CD56 cells and decreased the ratio of (CD4/CD8) cells [7].Dabrowski *et al*. exposed samples of mononuclear cells isolated from peripheral blood of healthy donors to 1,300 MHz; the results indicate that response of lymphocytes to phytohemagglutinin (PHA) as well as the T-cell suppressive activity (SAT index) and the saturation of IL-2 receptors significantly decreased in the culture supernatants. Also, the concentration of interleukin (IL)-10, IFNγ, TNF was significant decreased in the culture [8].

Material and methods

Subjects and methods:

Fourty five male volunteer's aged between 25 and 65 Years, for at least 10 years exposed to electromagnetic fields (EMFs) induced by transmission power lines in their residential area were used in this study . The EMFs was (with range 50 Hz) beside their homes this study carried out in three different cities of Baghdad included. (Al –Bladyat, Hay Al-Adel and Al-Dorra cities) during Feb. 2008, they were divided into three sub-groups according to distance from the towers of transmission power lines (1) range: from 1 to 25 meter (2) from 25 to 50 meter (3) from 50 to 75 meter.

Fifteen male of similar age. apparently healthy, with smoking habits unexposed, away from the transmission power lines about 500 meter was used as a control group. Five parameter were used for detection the effects of electromagnetic fields on the immune system included (CD 3, CD4, CD8, CD19, and CD56).

Isolation of Peripheral Blood lymphocytes:

Five ml of blood sample was drawn from each volunteer by vein puncture using disposable syringe containing 10-20 units of heparin/ml .Lymphocytes were isolated by density gradient sedimentation as described by Boyum .Blood samples were diluted 1:1 ratio in RPMI 1640 media , then layered over 2ml of Ficoll. Cooled centrifugation was carried out at 18 ºC for 20 minutes at 3000 rpm .Then the interphase was collected ,by Pasteur pipette, Ten µl of the cell suspension was applied to each well on immune fluorescence slides ,the slides were foiled with parafilm and kept at -20ºC until they used [9].

Immunostaining of lymphocytes:

The determination of lymphocytes phenotyping was performed by direct immune fluorescence technique as described by [10].

Ten µl of monoclonal antibodies includes (anti CD 3, anti CD4, antiCD8, anti CD19, and antiCD56), was dispensed over the spot in the slides. Counting of cells was performed using fluorescent microscope. A suitable countable field was located and the numbers of cells exhibiting fluorescence were counted, the calculation was made as follows;

$$\text{Percentage of labeled cells} = \frac{\text{Number of labeled cells}}{\text{Total no. of cells (200)}} \times 100$$

Measurement of ELF Frequency:

The electromagnetic fields were measured into three sub-sections according to distance (1) range: from 1 to 25 meter (2) from 25 to 50 meter (3) from 50 to 75 meter from the tower of transmission power lines. By using (Gauss Meter), in milli Gauss units, or micro Tesla (µT), 10 mG equals 1 µT.

Statistical Analysis

Experimental data were analyzed using statistical software SPSS 10.0 for Windows. Significance between control and samples was determined using Student's t-test. P value ≤ 0.05 was considered statistically significant.

Results

Mean percentage of isolated peripheral blood lymphocytes for the exposure volunteers groups and control group was illustrates in table (1).

The results demonstrate a significant reduction ($p<0.01$) in the mean percentage of CD3 cells (45.03%) ,In the all exposed volunteers groups to EMFs in all cities and in all distance in comparison to the mean percentage of control group CD3 (61. 35 %).as shown in figure (1).

A significant decrease ($p<0.01$) in CD 4 cells mean percentage (26.25%) was observed in exposed groups in all cities and in all distance as compared with the mean percentage of CD 4 in control group (42.72%) figure (2).

A significant decrease of mean percentage of CD21 cells ($p<0.01$) was observed (8.01%), In the exposed groups in all cities and in all distance in comparison with the CD21 mean percentage in control group (14.83%), figure (3).

The results indicated that the mean percentage CD8 cell was the only one that had not differed significantly in the study groups as compared to controls (26.72%), (25.41%) respectively, figure (4).

Significant reduction in mean percentage of CD 56 cells ($p<0.01$) was observed (7.22%) in exposed groups in all cities and in all distance as compared with the mean percentage of CD56 cells in control group (12.85%), figure (5).

CD4/CD8 ratio was of special importance because it presents an index that refers to the immunological balance between T-helper cells and T-cytotoxic cells in the immune system.

The result demonstrated that the mean percentage of $CD4^+/CD8^+$ ratio in the all exposure volunteers group (0.984 %)was significantly ($P<0.01$) lower in comparison to that in control groups(1.681%) as shown in figure (6).

Finally the result of measurement of electromagnetic field by Gauss meter for different site from the towers of transmission power line showed significant reduction with the distance, the values of Gauss meter reading reduced when the distance increased from the towers regression, and we can get the highest reading under the towers of power line 1.01, 0.987 ,and 1.142 μT in Al-Dora ,Hay al-adel and Al- Bldyat Respectively and the lowest reading 0.34 ,0.43, 0.61 μT in 75 meter away from the towers of transmission power line in Al-Dora ,Hayal-Adel, and Al-Bldyat Respectively. As shown in figure (7).

Table 1: Distribution of mean percentage of CD3+, CD4+, CD8+, CD21+ and CD 56+ lymphocytes in exposed volunteers and control group in the cities under the study.

CD marker	Distance meter	AL-BLDYAT	AL-ADEL	AL-DORRA	CONTROL
CD 3	0-25 m	46.2	41.8	38.3	61.35
	25-50 m	49.2	44.5	43.2	
	50-75 m	48.5	49.4	44.2	
CD 4	0-25 m	24.9	26.3	22.8	42.72
	25-50 m	27.2	26.3	25.4	
	50-75 m	28.3	28.7	26.4	
CD 8	0-25 m	27.2	26.6	25.4	25.41
	25-50 m	26.4	27.4	28.1	
	50-75 m	29.5	26.1	23.8	
CD 21	0-25 m	7.2	7.4	6.5	14.83
	25-50 m	9.8	8.5	8.4	
	50-75 m	8.4	7.1	8.8	
CD 56	0-25 m	8.5	7.1	5.4	12.85
	25-50 m	9.6	7.2	6.4	
	50-75 m	7.8	6.3	6.7	

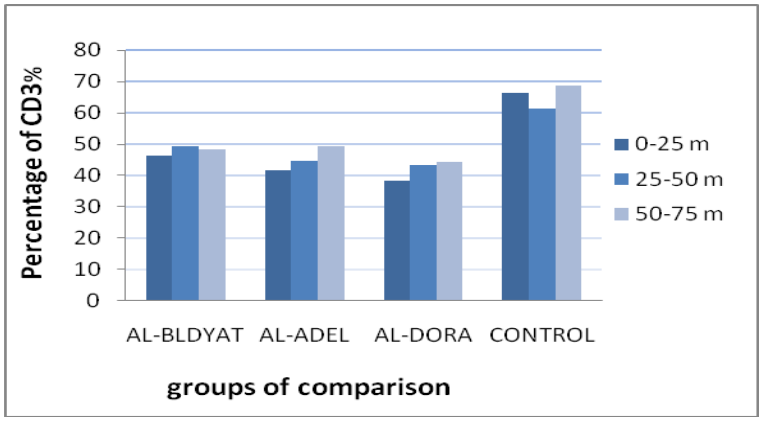


Figure1: the mean percentage of CD3 marker of PBL in all exposure and control groups with distance in meter

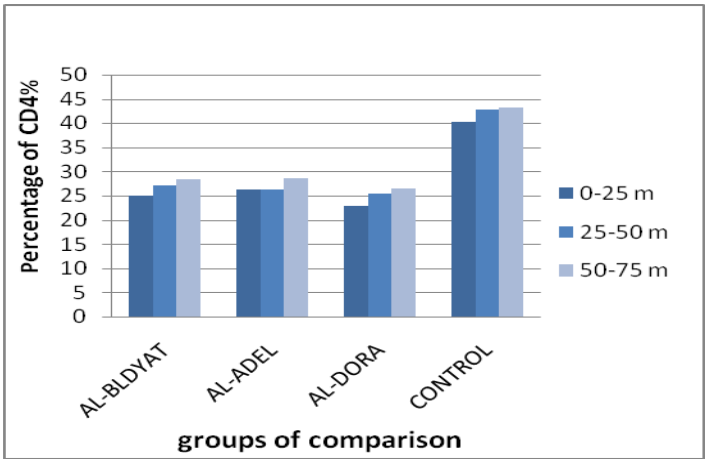


Figure 2: the mean percentage of CD4 marker of PBL in all exposure and control groups with distance in meter.

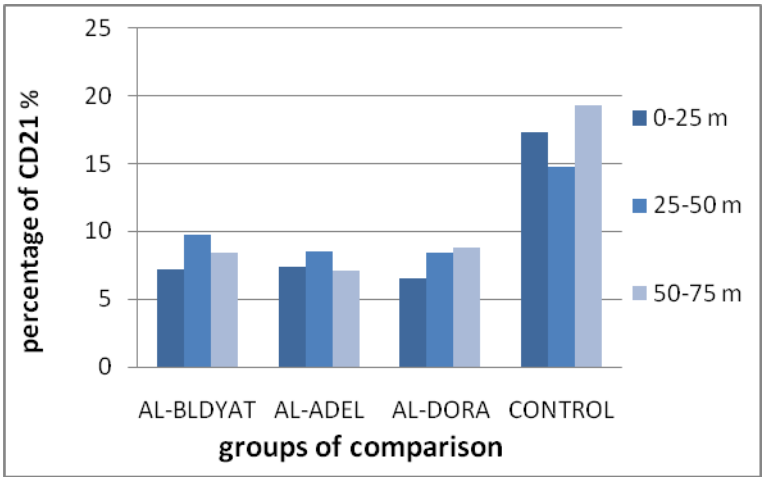


Figure 3: the mean percentage of CD21 marker of PBL in all exposure and control groups with distance in meter

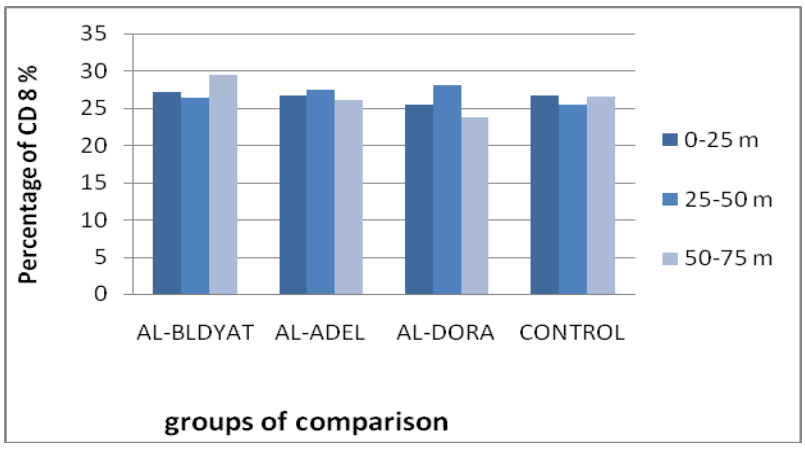


Figure 4: the mean percentage of CD8 marker of PBL in all exposure and control groups with distance in meter

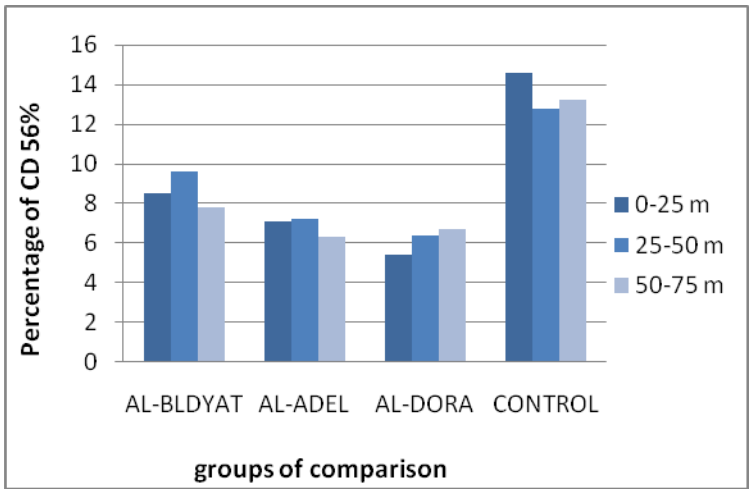


Figure 5: the mean percentage of CD56 marker of PBL in all exposure and control groups with distance in meter

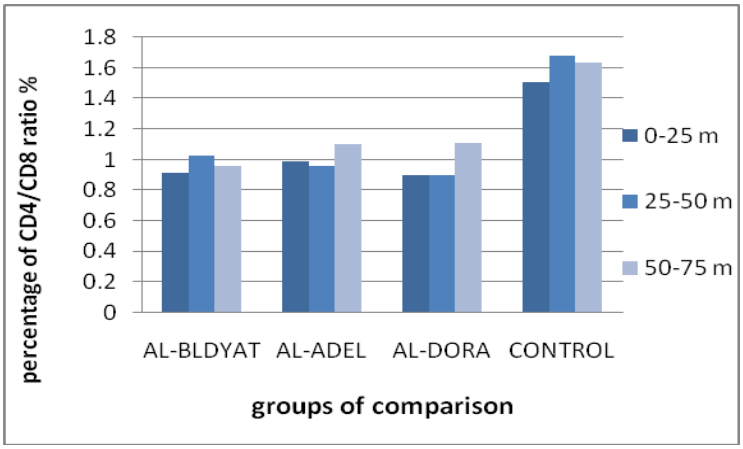


Figure 6: the mean percentage of CD4/CD8 ratio in all exposure and control groups with distance in meter

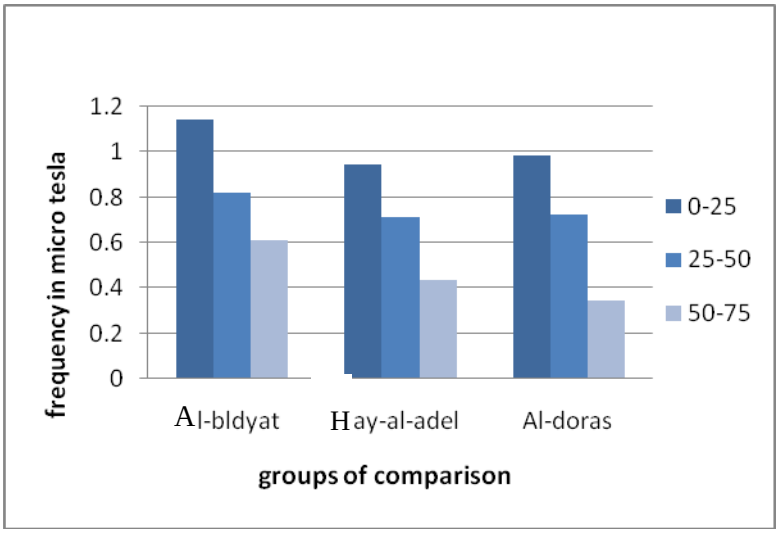
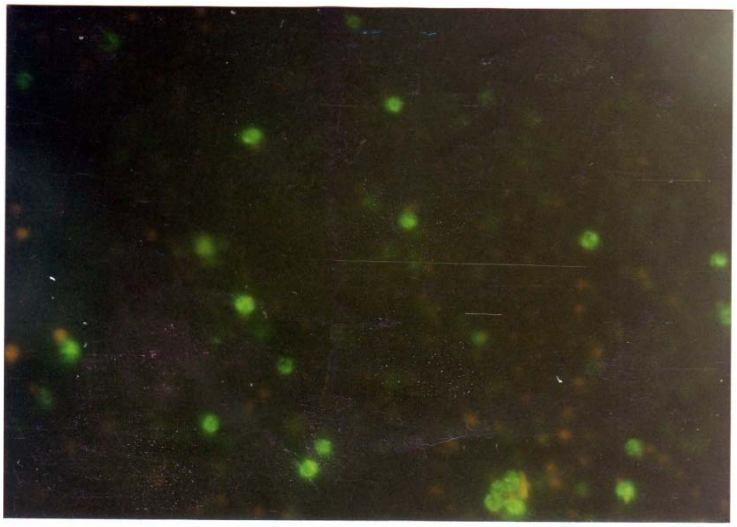


Figure 7: Measurement of EMFs in different site from the towers of power lines



A picture showed the lymphocytes subpopulation stained by direct immunofluorescence

Discussion

Surface CD markers of BPL are an important index for any research to clarify which immune defense mechanism is predominate and where are the weak points through which disease could have established [(11)].

The pivotal idea we can get from PBL phenotype is that all the exposure groups are immunosuppressed when we compared with the control group and this immune suppression which is reflected by low percentage of different PBL subsets could be result of the electromagnetic fields from transition power lines, as demonstrated by different recent studies [(12)].

Modulation signals are one important component in the delivery of EMF signals to which cells, tissues, organs and individuals can respond biologically. It is likely a key factor in determining whether and when

biological reactivity might be occurring [13].

We hypothesed that the frequency bands between 50 -60 Hz have been to alter immune responses and intercellular communication between lymphocytes such as altering the balance of cytokines, which regulate the growth of cells and determine whether the immune system will produce cells to proliferation and these reflected by the decrease in mean percentage of CD3, CD4, CD21, CD56, and the ratio of CDl4CD8 [14].

The cell membrane of lymphocytes contains docking ports on its surface called receptors that allow the cell to pick up distant chemical signals (cytokines, lymphokines ,hormones, neurotransmitters) sent by other Cells through the blood stream and local chemical signals generated by components of immune cells ,we think that many of these cell receptors also function as antennas for particular frequencies of electromagnetic fields and thus might lead to modulate the cell membrane receptor-mediated enzyme cascade in pre-B lymphocytes, with the implication and programmed-cell-death (apoptosis), cell cycle kinetics ,and cytokine expression [15].

Also exposure to EMFs may alter gene and/or protein expression in certain cell types, mRNA functions, immune responses and intercellular communication and this process may altered genes that responsible for the proliferation of different cell type of immune system such as Lymphocytes [16].

There is epidemiologic evidence that extremely low frequency (ELF $\leq$ 50 Hz) magnetic fields (MF) exposure associated with a decrease in melatonin production. (Melatonin is a hormone produced primarily by the pineal gland, located in the center of the brain), and this hormone has been found to protect cells, hemopoietic system tissues and organs against oxidative damage induced by a variety of free radical generating agents and processes[17] . According to this decrease damage to hemopoietic tissue, growth factors, cytokines, and genes involved in, apoptosis, signaling pathways and DNA repair might occur, which may decrease the percentage of lymphocytes subpopulations [18].

Several studies demonstrated that EMFs have genotoxic effects on human and animals. Significant increases in DNA damage including single and double strand breaks and cross-link chromosome conformation and micronucleus formation. Leaks in the membranes surrounding lysosomes could release digestive enzymes, including DNAase (an enzyme that destroys DNA). This explained the serious damage done to the DNA in cells by electromagnetic fields signals [19].

(EMFS) might interfere with regulation of the onset of differentiation and proliferation of B cell and apoptopic processes of actively proliferating cells. This mechanism reflects the low percentage of CD 21 and other subpopulation of lymphocytes [20] as shown in the results.

Finally we think that the chronic exposure to electromagnetic fields from power lines caused suppression in immune system which demonstrated in the results from the significant reduction in CD4/CD8 ratio as compared with this ratio in control group because the lower CD4/CD8 ratio the more immune suppression expected [21]. also we think that the more important mechanisms which caused immune suppression included DNA breaks in single or double strands and /or modulation signals

between immunological cells and cytokines whom responsible for the differentiation and proliferation of immune systems such as interlukines ,interferon ,Th1,and Th2.[22] .

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Ultra structural study of Carboxylester hydrolases activity in the interneuron of the mammalian spinal cord

Ali Abdul-Sattar Abdul-Rhman *PhD*.

Abstract

Background: Neurosciences mainly focused on the enzymatic pattern of the neurons, however, in this study the demonstration of certain carboxylaser hydrolases (Alph naphthyl butyrate esterases) activity in the Intrinsic spinal networks neurons (central pattern generator) was performed.

Objective: Interneurons or Renshow cells assume autoinhibitor functions, which dampens the a-motoneurons through negative feedback circuit, in addition to that they receive an input from higher centers through which it modify reflex responses to peripheral stimuli by facilitating or inhibiting different populations of interneurons ,however, this issue modulates the performence of specific movement performed by a-motoneurons.

Materials and methods: α – naphthyl butyrate used as a broad spectrum substrate in treating minced tissue block of gray matter of the spinal cord in the Rabbit prior to their traetment with the usual way to be examined lateron with the electron micrscopy .

Result: Reactivity was clearly evident in a form of deep stained granules in the nuclear matrex ,mitochondera and ,RER , and in certain preneuclear region ,however, the reactivity was varied in the mitochondrea in different neurons examined.

Conclusion: Rescentely the term, central pattern generators was used, which address the Intrinsic spinal networks of interneurones that control the timing and pattern of the muscle activity underlying locomotion in mammals,however the effect of the higher centers that modulates the type & tone of movement through those neurons elucidate function which was varied from excitatory-inhibitory, flexor-extensor. The reactivity of those enzymes and their different isoformes that migth have an effect on the molecular and genetic pattern of neurotransmitres were discussed in this study .

Keywords: Interneurons,Central pattern generator, Alpha naphthyl butyrate,esterase**,** Spinal cord

IRAQI J MED SCI, 2009; VOL.7 (2): 59-60

Introduction

Carboxylester hydrolases has been implicated in a wide range of novel dynamic functions which has been modulated according to the neuronal functions, development and regeneration[(1)].SinceThose are widely distributed in mammalian tissues, especially those with higher metabolic activity like brain, and spinal cord[(2)].

They are present as a complex tissue-specific mixture of various components, each of which presumably has a tissue-specific biological role [(3)]. However the metabolic function and the natural substrate of most carboxylester hydrolases in the different tissues are obscure [(4)].

Interneuron's assumes autoinhibitor functions, which dampens the α-motoneurons through negative feedback circuit, in addition to that they receive an input from higher centers through which it modify reflex responses to peripheral stimuli by facilitating or inhibition different populations of interneurons [(5)], however, this issue need profound

Dept. Human Anatomy, college of Medicine, Al– Nahrain University.
Adress Correspondence to: Dr. Ali Abdul-Sattar Abdul-Rhman
E- mail: col_med_nahrain@yahoo.com
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energy that can be served this tusk which translated in different forms of neurotransmitters and their precuersers in the stomata of those neurons.

The term used to highligths the physiological property of certain neurons in the gray matter of the cord ,which is the central pattern generators address the Intrinsic spinal networks that control the timing and pattern of the muscle activity underlying locomotion in mammals, howover, molecular and genetic approaches is a vital issue to elucidate the function of populations of CPG interneurons [6], the Execution of motor behaviors relies on circuitries effectively integrating immediate sensory feedback to efferent pathways controlling muscle activity [7]. It remains unclear how, during neuromuscular circuit assembly, sensory and motor projections become incorporated into tightly coordinated, yet functionally separate pathways. Since the early appearance of discrete intranerve trajectries suggests that trans-axonal interactions migth drive the segregation, within axial nerves, establishment of discrete afferent and efferent pathways depends on coordinate signaling between coextending sensory and motor projections. These heterotypic axon-axon interactions require motor axonal ephrin, guidance molecules which is one of the candidate molecules that performe this issue [8] .EphA3/EphA4 receptor tyrosine kinases activated by cognate sensory axonal ephrin-A ligands.Since genetic elimination of trans-axonal ephrin-A signaling in mice triggers drastic motor-sensory miswiring,culminating in functional efferents within proximal afferent pathways. Effective assembly of a key circuit underlying motor behaviors thus critically depends on trans-axonal signaling interactions resolving motor and sensory projections into discrete pathways.[9]

The α- motoneurons in the gray matter of the cord serive motor activities to the tonic and phasic muscle fibers showes different pattern of reactivites utilizing ANA esterases, and α- 1 and α-2 address the two varity of those neurons, is well established [10]. In the sence of this diversity ,the modulaters neurons must show different pattern in the neurotansmeters and macromolecules,synthsis and degradation which influence there differentation and activities .

Material & Methods

Carboxylester hydrolases was examined in the ventral horn cell of 10 New Zealand rabbits weighting 3-3.5 Kg. They were killed by sectioning of the great vessels of the neck without anesthesia. Laminectomy was performed, dura and arachnoid opened in the lumbo-sacral region. Cord segments with roots of sciatic nerve of both right and left sides were removed then placed in a small Petri dish containing 30 ml of saline solution at 37 Cº . Serial coronal sections with a sharp razor were cut through the ventral horn of the cord and then minced into a small slices of a bout 0.1cm cubes. Immediately, those slices were transferred to a small containers that contained a cold fixative composed of 2.5% glutaraldehyde buffered to pH 6.8 with 0.15M phosphate buffer for 2 hours at 4C, then blocks were further minced into a smaller pieces and washed (2-3) times with the same buffer prior to incubation. However some of blocks were processed without incubation in the substrate to be used as a control. Incubation medium was prepared according to the Modified after Bozdech & Bainton, 1981 [11], α- naphthyl butyrate (Fluka) 10mg (i.e.0.01ml), in

0.5ml ethanol. Phosphate buffer 0.15M pH 6.8. 9ml.and ,Hexazotized pararosanilin, 0.5ml , since this should be freshly prepared by mixing equal volumes of 1 ml 4% sodium nitrite (BDH), newly made, and 1 ml 4% pararosaniline (Fluka) in 2N HCl.

The cloudy mixture of the incubation medium was filtered with No.1 Whatman filter paper, prior to incubation which lasted for 2 hours, .and then washed overnight in phosphate buffer, treated with 1% osmium tetroxide for 1.5 hours, stained with 1% uranyl acetate, and embedded in Epon. Semi thin sections (0.5–1 μ) were obtained, stained with 1% methylene blue. Those sections were used for selecting the most adequate areas to be examined for the ventral horn neurons. Ultra thin sections (60–90 nm) were taken and examined in Philips CM10 electron microscope operating at 60 kV; some sections were examined without staining.

Results

Perfect results were obtained within 30 min of incubation in the ANB as the color of the minced tissues slices became dark - brown in color. Examination of the semi thin sections (0.5–1 μ) treated with 1% methylene blue, prior to be examined by electron microscope, delineated different sizes of ventral horn neurons. (Figure1)

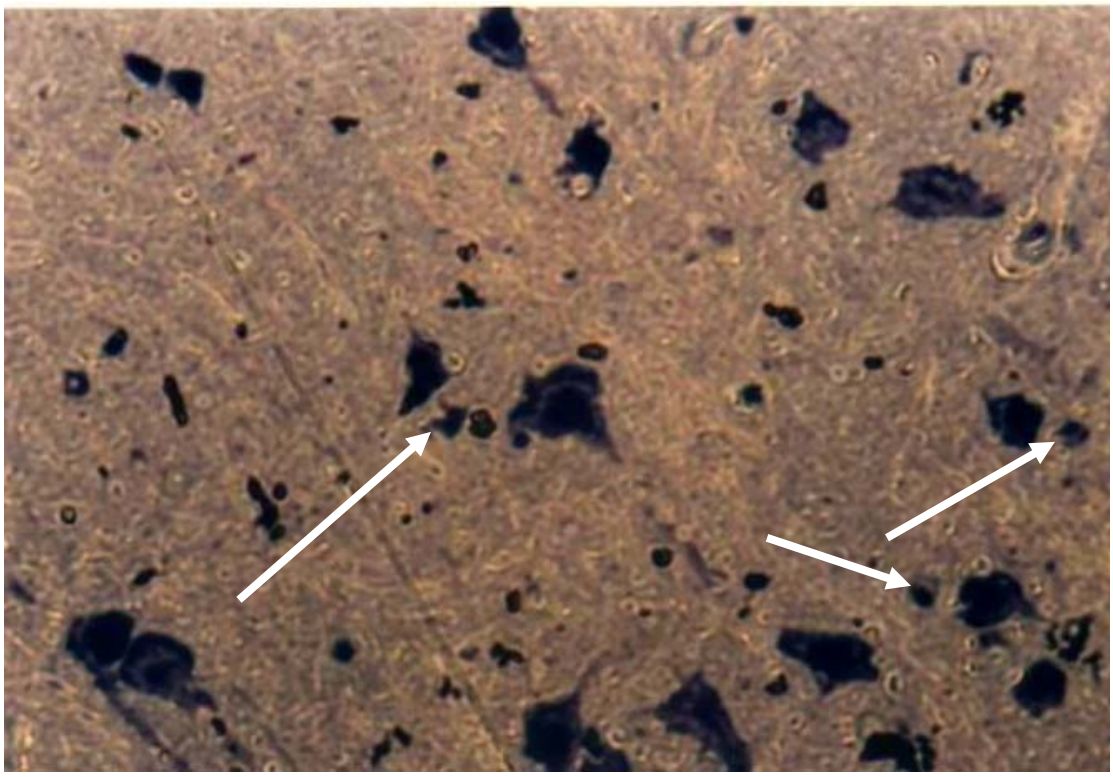

Figure1: Semi thin treated with methylene blue (X .400). Arrows interneurons

Small size neurons were selected for Electron microscopic examination which revealed that, those neurons are richly endowed with a variety of organelles that are loaded with black dots of the final reaction product (FRP). (Figure 2), conversely, sections from control blocks do not show this reactivity.

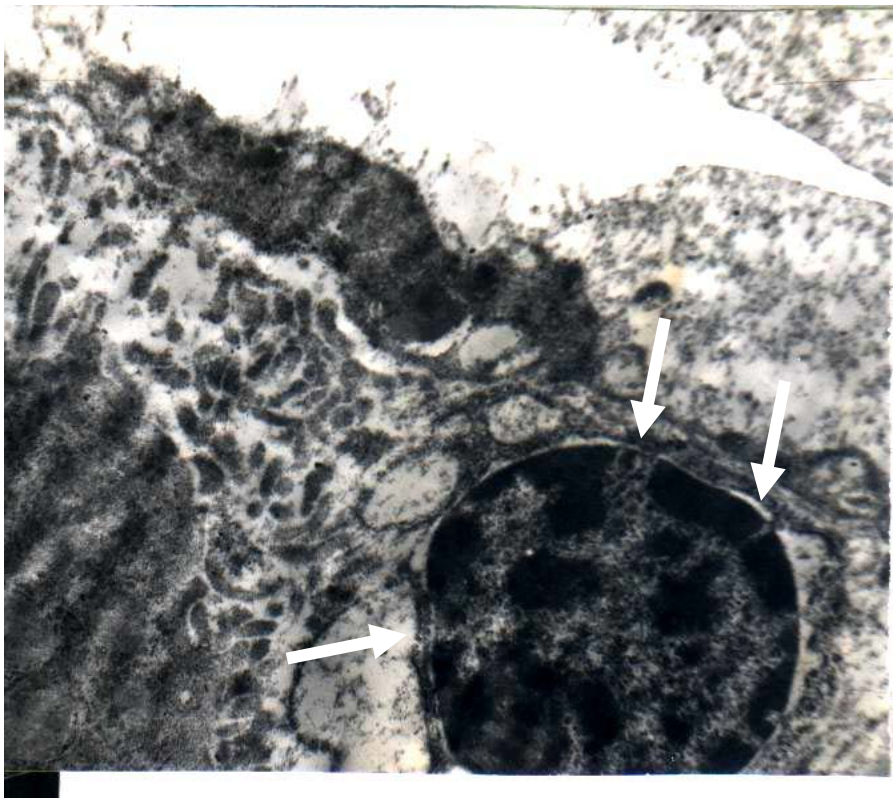

Figure2: Interneuron stained with ANBE. Arrows nuclear pores (X. 12000)

It is clearly evident that the nucleus of those neurons shows intense reactivity in the nuclear matrix with fenestrated nuclear membrane, it is difficult to identify nucleolus, as there are many that show their features regarding the size, since those are heavily stained.

With a higher magnification we overcome the failed of the somata, rough endoplasmic reticulum (RER) and mitochondria can be easily visualized with two forms of distribution in the cytoplasm. In (Figure 3). RER with their extensions to the Polyribosome were abundant, but mitochondria show varied in the intensity of the FRP from moderate to dark, although light ones have also been observed.

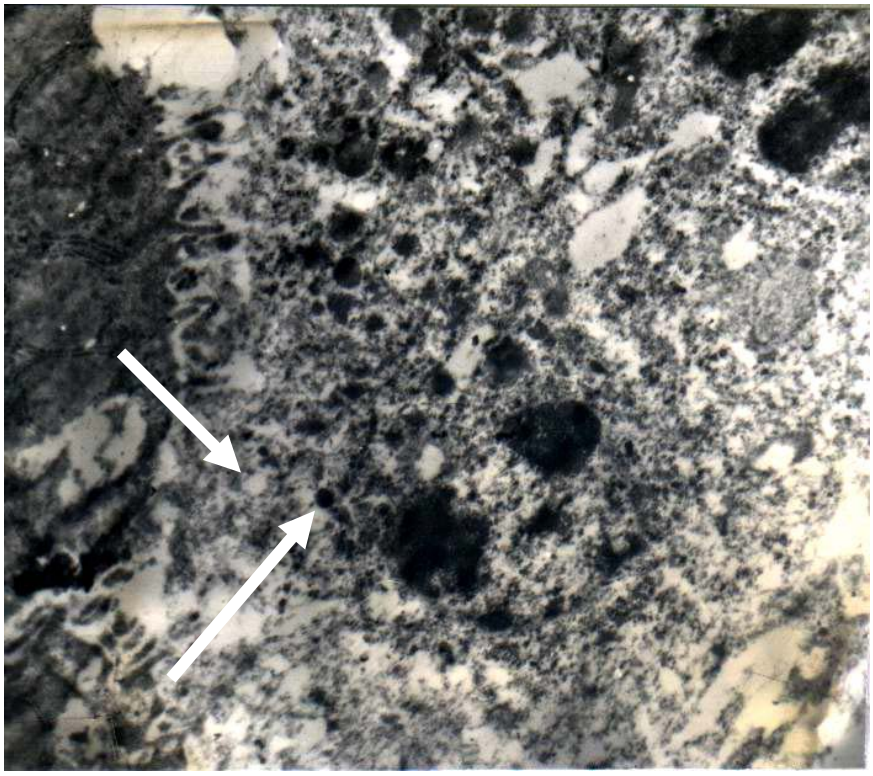

Figure 3: Cytoplasm of interneuron treated with ANBE .Arrows show mitochondria devoid from reactivity (X 24000).

While in other sections (Figure 4), the predominant intense staining was evident. In a form of a uniform heavily intense reactivity in the mitochondria (no light one have been observed) in addition to that the area surrounding the RER.

The main issue in the reactivity is that; the form of distribution of FRP is in the pre nuclear area not the neuropil (dendrites & axon terminals) as in some neurons,

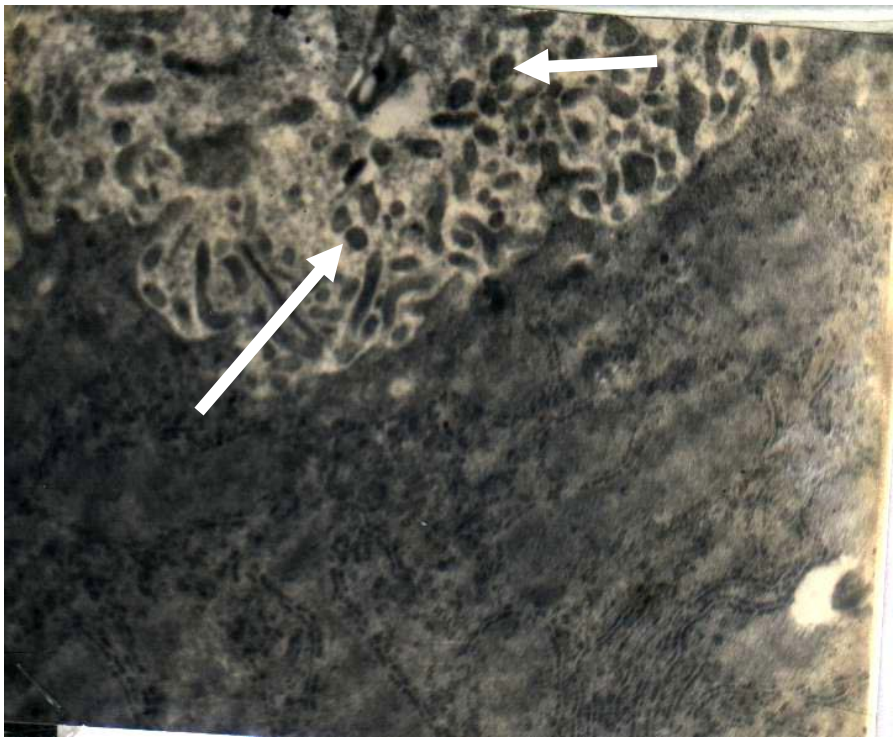

Figure 4: Cytoplasm of anther interneuron treated with ANBE .Arrows show mitochondria (X 24000).

Discussion

The spinal motoneurons are embedded within local neuronal; circuitries that determine stereotypic patterns of activities [12]. Presently, the internal organization of the mammalian locomotor central pattern generator (CPG) is unknown due to the difficulty in identifying and localizing interneurones involved in the network. The CPG was initially thought to be composed of half-centers, which set the basic locomotor rhythm by generating alternating excitation of antagonist motoneurone pools (e.gflexors and extensors) via reciprocal inhibition.[7].

Since the intrinsic spinal networks neurons control the timing and pattern of the muscle activity, through the α-motoneurons, however, central pattern generators neurons is a physiological term used to delineate those neurons in the mammalian spinal cord. The histochemical term in this aspect is in a limited in use for identification, as advances in transgenic technologies have greatly facilitated our understanding of the development and function of neural networks, many studies in this filed based on the overall structural role in rhythmic generation, organization of flexor-extensor networks, and the diverse role of commissural interneurons in coordinating left-right movements [6].

In this study we highlighted a histochemical glimpse on those neurons precisely in the lamina IX in the gray mater of the cord where the α-motoneurons existed. It's clearly evident from the shape, size and their locality in the vicinity to the α-motoneurons (Figure1), electron microscopical

reading indicates, tremendous activity of those neurons in the synthesis of transgenes for the different profile of carboxyl ester hydrolysis precursors regarding the ANB esterase, as utilized in this study which is visualized in the nucleus as huge black dots in the nuclear matrix, and the abundant nuclear pores in the nuclear membrane indicating active transportation of these precursors (Figure 2) [(13)].

The observation of Gallarda, et al 2008, about the segregation of axial nerve into motor and sensory fibers was based on the initial interactions between median medial column axon that extened to axial target and dorsal root ganglia neurites eventually resolve into sharply segregated proximal motor-sensory pathways,since the use of many polypaptide like nerve growth factor and neurotrophin-3, to select for nocioeptive and proprioceptive classes of sensory neurons,respectively ,they found that effective segregation of sensory and motor projections occurred irrespective of sensory subtype .Nevertheless motor axon of media medial column more frequently crossed into proprioceptive explants compared with nocioceptive cultures, homotypic (e.g. motor-motor) co-cultures failed to display axon segregation, stressing the heterotypic natuer of the underlying interactions.

On cellular bases the segregation of peripheral nerve fibers in to sensory and motor is well established due to the trans-axonal interactions [(14)].However axon-axon interaction have been implicated in olfactory and retinal axon targeting in Drosophila and mouse [(15)] .

To address the two forms of reactivity in the cytoplasm of interneurons on histochemical bases utilizing ANBE as a substrate (Figure 3 .4),this prompts the question of the two verities of interneurons ,in the cytoplasm precisely in the mitochondria (Figure 3) some of them were devoid from reactivity while in (Figure4) all the mitochondria shows reactivity.

The important issue is that, trans axonal reaction were inducted through the CPG neurons on α-motoneurons with two version in the reactivity of the ANB esterase which is one form of carboxylester hydrolases as, this form of carboxyl ester share in the synthesis and degradation of macromolecules formation in order to segregate motor axons that serve tonic or phasic muscle fibers. In this study it is an easy way in our histochemical practice to donate the trans-axonal interactions on motor nerve fibers with two forms of action modalities via ANB esterase, and CPG1, CPG 2 neurons in the gray matter of the mammalian cord is easily addressed via the activity of this form of carboxyl ester.

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A study for the correlation between eosinophil Derived Neurotoxin (EDN) and asthma.

Shehab.A.Lafei[1] *PhD*, **Nidhal Abdul-Mohymen[2]** *PhD*, **Amer Al-Najjar[3]** *PhD*.

Abstract

Background: Eosinophil Derived Neurotoxin (EDN) has been used to assess eosinophil cells activity and to monitor inflammation in asthmatic patients.

Objective: Study the correlation between Eosinophil Derived Neurotoxin (EDN) and asthma.

Materials and methods: Eosinophil Derived Neurotoxin (EDN) was extracted from eosinophils taken from patients with eosinophilic leukemia.This extract was conducted to study its biological effects (Gordon phenomena) and detection of antibodies against it in urine samples of diagnosed asthmatic patients.

Results: One of the two tested rabbits with the extracted EDN test material showed the signs and symptoms of Gordon phenomenon during the second day after injection and continued to show complete paralysis within the fifth day .Patients urine results showed that it contained a higher values of END than the control subjects urine .

Conclusion: Results revealed that EDN is a product that can be used as a monitor for asthma.

Key words: EDN, asthma, ELISA.

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Introduction

Pathophysiological changes of asthma are different , complex and involve interactions of several cellular and humoral mediators [1,2,3].Among the important cells types is the eosinophils.Four different cationic proteins and one neutral protein have been isolated from eosinophil. These products show potency to be a markers of the severity of inflammation [4,5].Major Basic Protein (MBP), constitutes the core of eosinophil granule and showed cytotoxicity to several helminthes, protozoa and bacteria [6,7]. It increases the hyperactivity on bronchial smooth muscle contraction in rabbits [8].

A positive correlation have been observed between the concentration of MBP and the number of thedesquamated epithelial in bronchoalveolar lavage as well as the degree of airway hyperresponsiveness in individuals [9].

MBP may induce airway hyperresponsiveness through its ability to inhibit binding of acetylcholine to muscarinic M2 receptors, resulting in the release of acetylcholine [10].Eosinophil Cationic Protein (ECP) , is a granule protein belongs to RNA–ase superfamily, it shows ribonuclease -3 activity. This protein is rich with arginine and its molecular weight is 18-21 KD [11].

ECP has many biological activities it elicits Gordon phenomenon when injected intrathecally in rabbits [12, 13]. It is able to release histamine from Mast cells [14]. Eosinophil Derived Neurotoxin EDN or eosinophil Protein x (EPX):-_It is a glycosilated single chain with protein with molecular weight of 18-21 KD. It is a member of ribonuclease -A-activity [13, 15]. Although EDN shows high sequence homology to ECP, it has

[1]Dept. Microbiology, college of Medicine Al-Anbar University, [2]Dept. Microbiology college of Medicine Al-Nahrain University, [3]Dept. Microbiology, college of Medicine Al-Al Mustansiyria University.

Adress Correspondence to: Dr. Nidhal AbdulMohymen.

E- mail: dr.nidhalmohammed@yahoo.com

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2000 fold greater ribonuclease activity than ECP and more neurotoxicity but not cytotoxicity [16]. EPX has been used to assess eosinophil activity and to monitor inflammation in asthmatic patients. [17].Oymer et al (2001) [18], found that atopic children have higher levels of urinary EPX than non atopic with acute asthma.

Asthma inflammatory Mediators can be detected in urine like histamine; leukotriens, eosinophil proteins (ECP, EDN) as well as interleukins like IL-8 [17, 19].There are different methods for the detection of different parameters and mediators useful in asthma diagnosis. Different clinical specimens are suitable for mediators detection such as Blood, Serum, Urine, Sputum and bronchoalveolar lavage. Variety of tests for asthma are available now, some of them are *in vivo* tests like skin prick test or *in vitro* tests as serological tests. So in this study we intended to study the presence of EDN in urine samples of asthmatic patients.

Materials and methods

Patients:One hundred asthmatic patients who attended the clinic of Allergy and Asthma in Ramadi General Hospital , Al-Anbar Governorate, during the period extended from July 2002 to January 2003. Patients were selected randomly from both sexes and their ages were from 10-50 years, and examined by specialized committee to be diagnosed as an asthmatics of different status, some of them were in rest and other were in attack. Skin test was done for all the patients.

Control Group: Twenty four healthy individuals from both sexes were selected resembling the same age groups of patients. They considered as negative control groups as they did not show a history of asthma and atopy after investigation.

Urine Samples:

Clean - catch, Mid – stream urine specimen was collected from each test and control individuals. They were advised to clean their external genetalia with soap and water, then the first stream was discarded and the Mid - stream urine sample (15 ml) was collected in clean dry sterile wide mouth

Eosinophil Derived Neurotoxin or Eosin

Eosinophil Derived Neurotoxin (EDN) was prepared from eosinophils isolated from blood sample withdrawn from eosinophilic leukemia patient as described below : Five milliliters of blood were taken from a patient suffering from eosinophilic leukemia attended Blood cancers center, Baghdad, Guide–lines of specimen collection were also regarded (WHO 1995). Blood film was done from the specimen as soon as possible and stained with Leishman stain and differential cell count was done it showed 45 % Eosinophils. Eosinophils were isolated and purified from blood specimen following method of Fernandez – Botran and Vetricka (2000)[20]: Five milliliters of blood in EDTA tube mixed with equal volume of 3 % dextran solution in normal saline . The mixture shaked gently and left for 45 minutes at room temperature and centrifuged for 5 minutes at 1500 rpm .The supernatant was discarded and the pellet was suspended in an equal volume of sterile normal saline to the starting volume of blood . This suspension was transferred into centrifuge tube and carefully underlayered with 3 ml of Ficoll solution 1.090 gr/ml gravity with clean and sterile syringe and centrifuged at 2000rpm at room temperature for 40 min.The mononuclear cell layer at the interphase was removed and the granulocyte pellet collected. This pellet

washed two times in Hanks balanced salt solution (HBSS).Using percoll powder, a percoll solution of the densities 1.090, 1.095 and 1.100 gr/ml was prepared in clean sterile glass centrifuge tube starting with solution of the lowest density (1.090 gr/ml).The granulocyte suspension as layered carefully on the gradient suspension, centrifugation at 3000 rpm for 30 min.The cell band at 1.095, 1.100 interphase was collected carefully with sterile and dry pasture pipette and washed with HBSS .Contaminating RBCS were lysed by cell pellet resuspension in 4.5 ml of ice cold water for 30 seconds and immediately, tonicity equilibrated with 0.5 ml of HBSS(free of Ca^{++} and Mg^{++}) .Eosinophil pellet was prepared from this suspension by centrifugation at 2000 rpm for 15 min, pellet kept frozen to be used for eosinophil granule extraction (Fernandez – Botran and Vetvicka 2000) [20] .

Release of Eosinophil granules: Eosinophil granules were released from the eosinophil Suspension obtained in the previous steps. Eosinophils pellet dissolved in 10 mls of sterile ice–cold 0.25 molar sucrose solutions. Cells washed once in this solution and precipitated by centrifugation at 2000 rpm for 15 minutes. Cells were resuspended in 10 mls of ice cold 0.25 molar sucrose and broken to release granules by repeated vigorous pipetting through a narrow–bore 10 ml calibrated pipette. Suspension was centrifuged at 10000 rpm in cold centrifuge for 20 minutes to sediment granules as a pellet [12]

Purification of Eosinophil Derived Neurotoxin EDN:Eosinophil granule pellet solubilized in 2 ml of 0.01 molar HCl($pH_{=2}$).This suspension centrifuged at 30000 rpm at $4C^{o}$ for 10 minutes to yield clear granule extract. The supernatant was chromatographed at 4 C^{o} on 1.2×47 cm sephadex G50 column equilibrated with 0.025 molar acetate buffers, pH 4.2. Flow rate was 12 ml/hr and 1.3 ml fractions were collected.Fractions eluting between 27 and 33 ml were pooled, dialyzed overnight against normal saline and centrifuged at 10000 rpm in cold centrifuge for 10 minutes.

Fraction (supernatant) was rechromatographed on a second sephadex G50 column equilibrated with phosphate buffer saline (pH 7.2).

Fractions eluting between 27 and 33 ml were pooled and concentrated on an Amicon filter for 2hours. Protein content of this concentrate was calculated using Lowry *et al*., 1951 (21) method. Concentrated extract was kept frozen at -20.

Identification of Eosinophil Derived Neurotoxin (EDN) by Polyacrylamide Gel Electrophoresis (PAGE): This test was done according to (12, 22).

Biological Test for EDN:

Local breed rabbits which were reared before experiment were used for this purpose. Two rabbits (each was weighing 1.5kg) were injected intracranially with sterile (0.25 ml) of EDN extract containing (250 μg) of protein. Another two rabbits were used as control injected with (0.25 ml) sterile normal saline in the same route.

Intracranial injection of the test and control material was done under high precaution and carefully using surgical theater of small animals in the College of Veterinary Medicine, Baghdad University. The site of injection was prepared properly by clipping and shaving the site of operation, washing with soap and water and disinfections with tincture iodine 2.5%. A burr hole was done in the skull, using sterile stainless steel trephine and injection of (0.25 ml) of the test material was done into the right occipital cavity. The skin closed with sterile silk as single interrupted suture.

The same thing was done for another two rabbits as control, injected with 0.25 ml sterile normal saline (Figure 1).

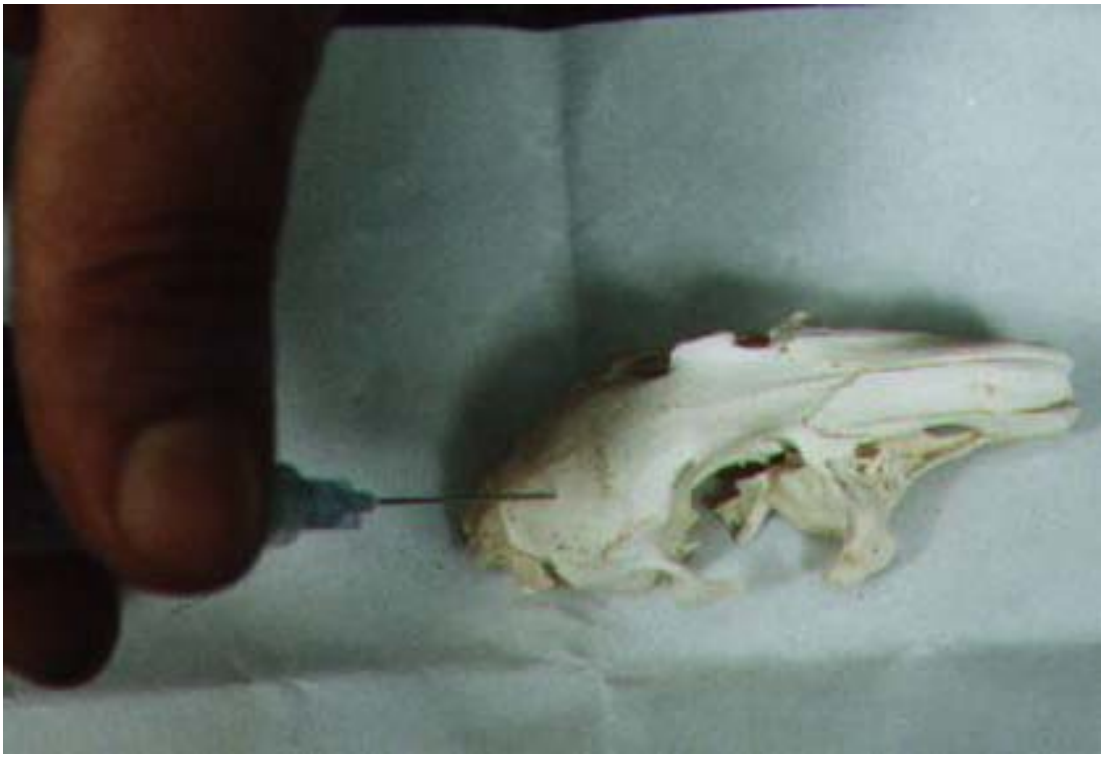

Figure 1: Skull of rabbit to show the site of intracranial injection

Stitches were removed after eight days and animals observed for 10 days [13].

Detection of EDN Antibodies in Urine Samples: Enzyme linked immunosorbent assay (ELISA) test was used for EDN detection in urine samples as described bellow: After 10 fold dilution of extract (EDN) with sterile coating buffer, 100ml of EDN solution was dispensed in each well of the microtiter plate except well A .The plate kept at $4C^{o}$ (in the refrigerator) for overnight. And the test was completed according to (20)using Rabbit Antihuman IgG conjugated with peroxidase, Bio–kit) diluted in specific dilution solution was added to each well except well D. Cut– off value was calculated following method of (AL-Murrani et al 2000) [23].

Statistical Analysis

Data were analyzed using chi square and cross tabulation, following methods of (Daniel 1999) [24] and computer type Pentium4.

Results

One of the two tested rabbits with the extracted EDN test material showed the signs and symptoms of Gordon phenomenon during the second day after injection and continued to show complete paralysis within the fifth day (Figure 2).

Symptoms of Gordon phenomenon were: stiffness mostly in

the fore limbs and dropped to the floor. Complete ataxia later and the animal showed good appetite. Animals were injected with sterile normal saline did not show any sign or symptom or even any abnormality due to post–operative complications (Figure 3).

Figure 2: Positive Gordon test

Figure 3: Negative Gordon test in control rabbit

Detection of extracted EDN by PAGE showed separation of a single band in the same level (line) of the β-lactoglobulin band. This means that the separated test band was a protein of the molecular weight nearly equal to that of the molecular weight of the indicator protein (β-Lactoglobulin, 1800 Daltons).

Trypsinogen band was seen about 6mm before the two above mentioned bands (Figure 4).

Figure 4: Distance traveled by test and Marker proteins IN PAGE.

ELISA test for EDN in Urine

Results showed that there was no significant difference($p > 0.05$) between absorbance mean values for EDN in urine of positive and negative sputum culture groups, (both in attack and rest status of asthma). Control samples showed lower mean values than that of test groups (Table 1).

Table 1: ELISA readings for EDN in urine of the studied group

Age and sex		Attack				Rest			
		10-17year		18-50year		10-17 year		18-50 year	
		Male	Female	Male	Female	Male	Female	Male	Female
Positive sputum	mean	0.081	0.074	0.0907	0.12	0.062	0.076	0.080	0.084
Negative sputum	mean	0.074	0.066	0.093	0.069	0.088	0.076	0.071	0.094

Cut off value =0.050

Discussion

Polyacrylamide gel electrophoresis of eosinophil derived neuroprotein (EDN) extracted from eosinophilic patient showed one band with molecular weight approximately to 18000 Daltons, this result was nearly similar to that gained by (13,15).

Extracted EDN showed Gordon phenomenon in one of the two tested rabbits and this was lower than that of (12), this difference might be due to: Purity of extracted EDN, difference in the breed of the rabbits (in this study local breed rabbits was used) which might be less sensitive to EDN than the New Zeland rabbits were used by (13).
Results obtained showed no significant difference between absorbance mean values of EDN in urine of patients with positive culture and that of patients with negative sputum culture, this might be due to the efficacy of allergens and infective agents in both groups to induce EDN release via eosinophil activation [19]. A number of studies have indicated that the assessment of eosinophil-derived proteins in various body fluids could be used for monitoring disease activity of asthma. Due to the relationship between levels of eosinophil proteins in serum/urine samples and lung function, as well as significant concentration differences between symptomatic and asymptomatic asthmatics, the assessment of eosinophil proteins in serum or urine samples appear to be more appropriate in monitoring disease activity [17, 18].
Different studies mentioned that EDN may be considered as an inflammatory mediator for many reactions in addition to asthma such as endocarditis, entritis and dermatitis [25, 26, 27].

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Nerve Conduction Studies in Healthy Iraqis: Normative Data

Farqad B. Hamdan *MBChB; MSc; PhD.*

Abstract

Background: Nerve conduction studies as part of the peripheral neurophysiologic examination are an extension of the clinical history and examination. They can be extremely useful both in localizing lesions and determining the pathological processes.

Objective: To establish the normal electrophysiological data for the facial nerve and commonly tested nerves of the upper and lower limbs in healthy Iraqis and to compare with those data published in the literature.

Methods: Nerve conduction studies were performed in the upper and lower limbs of 11,437 carefully screened healthy individuals between the ages of 2 months and 89 years using a standardized technique.

Results: The data were separately analyzed for different age groups. In the age group less than 10 years, the sensory and motor nerve conduction velocities were progressively increasing with increasing age until adult values were reached. Later, in the adulthood, the conduction velocity of all tested nerves decreased with age and this is pronounced in both lower and upper limbs.

Conclusion: Normative conduction parameters of the facial nerve and peripheral nerves in the upper and lower limbs were established for the EMG laboratory in Iraq. The overall mean sensory and motor nerve conduction parameters for the tested nerves compared favorably with the existing literature data.

Keywords: Upper limbs, Lower limbs, Nerves, Conduction Velocity, Iraqis

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Introduction

Nerve conduction studies (NCS) afford data on peripheral nervous system function which may be used to provide diagnosis, description of disease state (old/new; dynamic/static pathophysiology), longitudinal monitoring of disease by using multiple studies, and advice on prognosis and management based on the test results and the disease detected [1-6].

Another type of NCS is referred to as late response (F-wave testing) and is usually performed on nerves more proximal to the spine.

Dept. Physiology, college of Medicine Al-Nahrain University.
Adress Correspondence to: Dr. Farqad B. Hamdan
E-mail: farqadbhamdan@yahoo.com
Baghdad, Iraq, Tel. + 9647901658795, P.O. Box 14222.
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These segments include the first several centimeters of a compound nerve emerging from the spinal cord or brainstem.They are helpful in diagnosing conditions of radiculopathies, plexopathies, polyneuropathies, and proximal mononeuropathies [6]. Late response studies are additional studies complementary to nerve conduction velocity and are performed during the same patient evaluation [1].

Age-matched "Normal" values for NCS parameters are either derived from studies of groups of neurologically normal subjects or culled from the literature [7]. Many studies have been published from the Western countries regarding normative data for the nerves of the upper and lower limbs [8-13]. Unfortunately, in the Middle East, such studies are limited; in the best conditions, the number does not exceed few hundreds [14-17].

For the last few decades, electrophysiology laboratories have been applying standard values used by the US, Canada and Europe to diagnose different neurological problems.

This study is therefore intended to obtain a set of data from a large scale of healthy Iraqis in order to establish reference values for the local EMG laboratory and to compare Iraqi values with worldwide published data.

There are a number of physical parameters that require correction or allowance for when establishing normative electrophysiological data. The most important is temperature i.e., the motor nerve conduction velocity (MNCV) is reduced by approximately 1 m/s per °C temperature fall. Some measures of conduction require correction for limb length or height. Finally nerve conduction data alter with age. The motor conduction slows by 0.4–1.7 m/s per decade after 20 years and the sensory by 2–4 m/s [(7)].

This paper provides normative electrophysiological data for the facial, median, ulnar, radial, musculocuatneous, axillary, femoral, common peroneal, and tibial nerves in healthy individuals at different age groups using standard distances and temperature control.

Subjects and Methods

Eleven thousands and four hundred thirty seven healthy individuals, aged 2 months to 89 years (36.39±15.36), were included in this cross-sectional study. They comprised 6,325 women with an average age of (38.05±13.79 years) and 5,112 men with an average age of (34.11±17.04 years).

Two thousands and four hundred forty four subjects were specifically recruited as normal. The rest were felt normal, so an informed consent was taken from them or their parents or relatives to do multiple nerve conductions on the normal limb(s).

The data were collected from individuals attending the EMG unit at Al-Kadhimiya Teaching Hospital, Baghdad-Iraq for neurophysiologic assessment during a 14-year period (1992-2006). All individuals were screened for inclusion criteria that comprised normal neurological physical examination, normal laboratory findings regarding serum sugar, electrolytes and renal function. A standardized questionnaire was used to exclude those with a history of systemic or neuromuscular diseases.

The following individuals were excluded: those with a history of alcohol abuse or medications that might affect the results, and those with a history of diabetes, hypothyroidism and systemic diseases. None of the individuals were taking any medication at the time of conducting the EMG study.

A basic neurological examination was performed to assess muscle power, stretch reflexes and sensations both superficial and deep. Room temperature was maintained at 25 °C. The EMG study was performed with the subject lying comfortably in the supine position. A standardized technique was used to obtain and record action potentials for motor and sensory functions [(18)].

A 4-channel electromyography machine (Dantec, Denmark) was set as follows: for motor nerve conduction, the low cut filter was 2–5 Hz and the high cut was 10 KHz. For sensory nerve conduction, low cut: 5–10 Hz, high cut: 2–3 KHz; the amplification between 20,000 and 100,000 times; electrode impedance was kept below 5 kΩ; sweep speed for sensory nerve conduction: 1–2 ms/ division while for motor nerve conduction: 2–5 ms/ division; and a

stimulus duration of 50 μs to 1000 μs and current 0–100 mA are required for effective nerve stimulation. Supramaximal stimulation (10%–30% more than the current required for maximal action potential) was used.

Data were collected for the following parameters: distal latency measured from the onset of action potential, conduction velocity, and amplitude of compound muscle action potential (CMAPA) and sensory nerve action potential (SNAPA) were measured from positive peak to the negative peak.

The following nerves were evaluated: for motor nerve conduction velocity– facial, median, ulnar, radial, axillary, musculocutaneous, tibial, common peroneal, and femoral nerves; for sensory nerve conduction velocity (SNCV) – median, ulnar, radial, musculocutaneous, tibial, and superficial peroneal nerves were evaluated.

The motor nerves were examined orthodromically and stimulated with bipolar surface stimulating electrode (Dantec 13L36) at two points along its course. The action potential was recorded with a concentric needle electrode (Dantec 13L50) placed close to the motor point of the muscle. A ground electrode (Dantec 13S93) was placed between the stimulating and recording electrodes. The MNCV was calculated using the distance between points of two stimulations by latency of that segment.

Stimulation of facial nerve was achieved at a point just below the ear and anterior to the mastoid process. Its motor response was recorded from orbicularis oris muscle. Distal median and ulnar stimulations were performed 8 cm proximal to the needle electrode. The site was medial to flexor carpi radialis tendon for the median nerve while posterior to flexor carpi ulnaris tendon for the ulnar nerve. Proximal median nerve stimulation was performed medial to biceps brachii tendon at the elbow crease. For ulnar nerve, proximal stimulation was just distal to the medial epicondyle of the humerus with the elbow in 70° flexion. Recordings of the muscle response for the median and ulnar nerves were done from abductor pollicis brevis and abductor digiti minimi muscles, respectively.

The radial nerve was stimulated distally at the lateral edge of extensor carpi ulnaris muscle, 8 to 10 cm proximal to the styloid process of the ulna and the recording from extensor indicis muscle. Proximally, radial nerve was stimulated between brachioradialis and the tendon of biceps brachii 6 cm proximal to the lateral epicondyle of the humerus. The musculocutaneous and axillary nerves were stimulated at the posterior cervical triangle 3 to 6 cm above the clavicle just behind sternocleidomastoid muscle. Muscle action potentials were recorded from biceps brachii and deltoid muscles, respectively.

For deep peroneal motor nerve conduction evaluation, the recordings were obtained from extensor digitorum brevis. The stimulation was given at the ankle distally and 2 cm distal to the fibular neck (below knee) proximally. Motor conduction study of the tibial nerve recorded the muscle response from abductor hallucis muscle after stimulation at the popliteal fossa and at the ankle posterior to the medial malleolus. The femoral nerve was stimulated below the inguinal ligament and lateral to the femoral artery. Motor conduction was recorded from rectus femoris muscle 14 and 30 cm distal to the point of stimulation.

Sensory nerve conduction was measured antidromically. The SNCV was measured by stimulating at a single site. The sensory conduction velocity was calculated by dividing the distance between the stimulating and the recording sites by latency.

The active ring electrode (Dantec 13L69) was placed over the 1st, 2nd and 5th digit to record responses along the radial, median and ulnar nerves, respectively. The reference electrode was placed about 4 cm distal to the active electrode. Median nerve stimulation was performed at 13 cm proximal to the active electrode and medial to flexor carpi radialis tendon. For the ulnar nerve, stimulation was performed 10 cm proximal to the active electrode and posterior to the flexor carpi ulnaris tendon. Radial nerve stimulation was performed 10 cm proximal to the active electrode along the lateral border of the radius.

The musculocutaneous was stimulated between the tendon of biceps brachii medially and brachioradialis laterally. The Self adhesive disposable surface recording electrode (Dantec C13L20) was placed 12 cm distal to the point of stimulation over the course of the nerve in the forearm and along the straight line from the stimulus point to the radial artery at the wrist.

Sensory conduction studies of the tibial nerve (medial planter nerve) from the first toe with ring electrodes and antidromic stimulation at the ankle (at the same site that elicits the motor response). The superficial peroneal nerve was stimulated against the anterior edge of the fibulae 12 cm proximal to the active surface electrode located just medial to the lateral malleolus at the ankle.

Because of difference in length of the upper and lower limbs among individuals and thus differences in length of the reflex arc, the disparity was taken into account. In order to quantify individual differences, the distance between the anterior superior iliac spine and the lateral malleolus for the lower limbs and the distance between the seventh cervical spinous process and the wrist for the upper limbs were considered.

Data were processed using the Statistical Package for Social Sciences (SPSS). Descriptive statistics for continuous variables included the mean and the standard deviation. Correlation coefficient (r) was used (P value < 0.05) to assess the relation between the age of the subjects and the conduction velocity of the nerves studied.

Results

Data were separately analyzed for each age group. In the age group less than 10 years, the SNCV and MNCV of median, ulnar, radial, tibial, and common peroneal nerves progressively increased (positively correlated) with increasing age until they reached adult values. Late in the adulthood (age groups 60 years and above), the conduction velocity of all the nerves tested decreased with age (negatively correlated).

The mean and standard deviation for the tested nerves are summarized in tables 1 through 9. Tables 10 through 19 show a comparison between the results of the present study and those reported by others worldwide.

Discussion

This study examined nerve conduction parameters of multiple nerves in the upper and lower limbs of a

healthy sample in Iraq under standardized conditions. The aim is to provide normative and reference values for Iraqi EMG labs. A comparison was made between the results obtained in this study and other published data with a special emphasis on studies that used standardized techniques and recorded the age of the subjects [8, 9, 13, 14, 16, 17, 19-39].

Age has been widely accepted to have an influence on nerve conduction velocity. Consequently, many laboratories have produced normative nerve conduction velocity values which are divided according to age groups. Many investigators have attempted to study the association between aging and nerve velocities, both motor and sensory[40-44]. In this study, the statistically significant increment or decrement of NCV with age collaborates with the findings of other researchers [45-49].

The motor parameters of the facial nerve as recorded in this study generally coincide with the results of other researchers [19-21]. The motor latency of axillary nerve was in close proximity to that reported by Kraft [22]. The SNCV values of musculocutaneous were close to the values reported by other workers [23-25], while the SL and SNAPA values were less than the reported values by Kraft [22] and Spindler and Felsenthal [25] (table 10).

For the femoral nerve motor latency, our data were in close proximity to those reported by Uludag et al. [15] and Gassel [26]; whereas the DML and MNCV are far less than that reported by the latter worker and Johnson et al. [27] (table 10).

For the median sensory parameters (table 11), the SNCV is less than that reported by Hennessey et al. [8] and Karagoz et al. [28] but similar to the results of Shehab [14], Awang et al. [16] and Kimura [18]. Moreover, the recorded SNAPA is less than that reported by Shehab [14] and higher than that of Hennessey et al. [8], Kimura [18], and Karagoz et al. [28].

On the other hand, the results of the motor parameters were more favorable with the almost similar MNCV results of others [8, 14, 16, 18, 28]. The median nerve CMAPA in this study was similar to that of Hennessey et al. [8] and Shehab [14], but higher than that of Kimura [18] and Karagoz et al. [28].

The data of ulnar SNCV were in agreement with the findings of Shehab [14] and Kimura [18] but less than those reported by others [16, 27]. Ulnar SNAPA was similar to the results of Shehab [14] but rather higher than that of other researchers [18, 28]. The motor data of ulnar nerve showed a good similarity with those reported by Shehab [14], Kimura [18], Karagoz et al. [28], and Buschbacher [29]. Apart from this, the MNCV was higher as compared to that of Awang et al. [16] and the CMAPA was also higher in comparison to that of Shehab [14] and Kimura [18] (table 11).

Concerning the results of radial nerve (table 11), the SNCV, MNCV, and CMAPA values were in close proximity to the findings of Shehab [14], Jebsen [30], and Trojaborg and Sinrup [31]. On the other hand, the SNAPA was higher than that reported by Falco et al. [9], Johnson et al. [13], Shehab [14], and Trojaborg and Sinrup [31].

In table 12, it can be observed that the sensory and motor CV of the tibial nerve was favorable with the results of Kimura [18], Buschbacher [32], Budak [33], and Antunes et al. [34]. However, the SNAPA and CMAPA were higher than that reported by Antunes et al. [34] and Kimura [18], respectively. On the contrary, the CMAPA was less than that

reported by Buschbacher [32] and Antunes et al. [34].

Regarding the data of common peroneal nerve, SNAPA, MCNV, and CMAPA values were similar to those encountered by Kimura [18], Karagoz et al. [28], DiBenedetto [35], Buschbacher [36]. However, the SNCV and MNCV were slightly lower than that reported by DiBenedetto [35] and Karagoz et al. [28], respectively (table 12).

As presented in table 13, the values of F-wave parameter recorded from the median, ulnar, tibial, and common peroneal nerves were in accordance with those reported by Kimura [18], Alavian-Ghavanni and Haghparah [17], Budak [33], Buschbacher [37-39].

To sum up, the values for most of the nerves tested agree with most of the other researches while few nerve parameters were showing a considerable departure. The difference between the results of the present study and the data published in the literature could be attributed to variety of causes.

Firstly, the difference in the distance between the stimulating and recording electrodes and the muscles tested which inflicted well on the lower values of median and ulnar SNCV and femoral MNCV reported in this study i.e., the latter nerve was tested from the rectus femoris muscle while it was studied by Johnson et al [27] from vastus medialis muscle. .

Secondly; the age of the subjects studied. Most studies used middle aged subjects (between 20 years and late fifties) [14, 16, 29, 32, 36-39] while we extended the study to include younger groups (those below 1 year) and older groups (above 80 years). Consequently, the effect of age on conduction velocity was more clearly evident in the present study (tables 1 through 9).

Thirdly; number of the subjects examined. In the best conditions of other studies, the number of examined subjects does not exceed few hundred [14-17]. This number is far less than the number of subjects included in the present study. It is doubtless that increasing the number of subjects examined will smooth the data and reduces the bias in the statistics.

Fourthly; the diversity of the methods and techniques (studies differ in maneuvering, setting, recording the electrical response, and equipment used). The SNAPA for the nerves tested in this study was higher than the data published in the literature because we measured the amplitude from the peak of the negative potential to the peak of positive potential rather than from the baseline to the peak of the negative potential as adopted by others [8, 9, 13, 14, 18, 28, 31, 34, 35].

The type of electrode used could also be a source of variation. Surface electrodes as used by Shehab [14], Awang et al. [16], and Buschbacher [29, 32, 36-39] are designed to give information about the whole muscle stimulated. Such electrodes will record the time taken for the fastest axons to conduct an impulse to the muscle.

In the present study where a needle electrode was used, accurate conduction time information were obtained even with simultaneous activation of many nerves[22]; nevertheless, needle electrodes record from only a small area of muscle or nerve, which necessarily provide more complex information and making numerical analysis difficult [7]. Yet, NCS are routinely performed with needle electromyogram rather than surface electrodes. The routine use of needle electrodes enables the examiner to determine the site and extent of peripheral nerve pathology [2-6]. Providing standardized data using a

needle electrode is more essential from the practical point of view.

Finally; the ethnic group studied. Some studies were done on Caucasian subjects, others on Asian; however, none of the studies examined the effect of age on NCV among Caucasian populations living in different geographical areas. Similarly, there is no study comparing the effect of aging on NCV between Iraqis and Asian.

At present it is difficult to attribute the differences to a single factor. On the other hand, the diversity could have resulted for variables that were not yet considered by workers such as body built and climatic dwelling conditions. Further clarification will wait studying the effect of factors such as body mass index on NCV.

Conclusion

Normative conduction parameters of facial and peripheral nerves in the upper and lower limbs were established for our EMG lab in Iraq. The overall mean sensory and motor nerve conduction parameters for the tested nerves compared favorably with the existing literature data.

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Table 1: Facial Nerve Parameters in Different Age Groups

Age Group (years)	DML (msec)	MNCV (m/sec)	CMAPA (mV)
1-10 (n = 176)	3.03±0.1	24.88±1.78	1.4±1.8
11-20 (n = 203)	2.97±0.3	31.02±1.8	1.8±1.2
21-30 (n = 288)	3.06±0.19	30.17±2.11	2.1±1.9
31-40 (n = 356)	3.18±0.22	33.3±3.19	2.8±2.1
41-50 (n = 477)	3.3±0.47	31.44±3	3.0±2.1
51-60 (n = 291)	3.27±0.71	29.4±4.06	3.1±2.4
61-70 (n = 178)	4.1±0.91	28.9±5.33	2.6±1.9

The values are presented as mean±SD, n = number, DML = distal motor latency measured from the onset of action potential, MNCV = motor nerve conduction velocity, CMAPA = compound muscle action potential amplitude measured from peak to peak.

Table 2: Median Nerve Parameters in Different Age Groups

Age Group (years)	SL (msec)	SNCV (m/sec)	SNAPA (µV)	DML (msec)	MNCV (m/sec)	CMAPA (mV)	F-wave (msec)
< 1 n = 442	1.37±0.15 (1.2-1.5)	35.37±6.65 (30.0-42.8)	36.1±8.4 (13.6-54.8)	2.83±0.4 (2.4-3.2)	46.83±2.78 (44.8-50.0)	9.12±4.32 (5.88-15.2)	15.97±1.56 (14.5-17.6)
1-10 n = 678	1.53±0.25 (1.2-2.4)	52.8±4.71 (42.3-59.1	57.3±15.2 (35.2-73)	2.61±0.59 (1.8-3.6)	56.35±5.03 (44.0-61.5)	13±7.75 (8.8-24.6)	21.7±3.80 (17.2-28.8)
11-20 n = 716	1.84±0.23 (1.5-2.5)	53.35±3.75 (45.0-61.0)	57.8±17.2 (20-67)	3.28±0.47 (2.0-4.2)	58.73±5.21 (48.9-68.2)	13.98±3.5 (9-17.1)	26.89±2.36 (20.6-32.0)
21-30 n = 1110	1.87±0.18 (1.5-2.5)	52.98±3.83 (43.5-66.6)	61.1±29.57 (14-140)	3.34±0.45 (2.3-4.8)	59.72±4.39 (47.7-68.5)	15.83±5.57 (7.7-30)	26.67±2.31 (21.3-35.4)
31-40 n = 1301	1.94±0.2 (1.5-2.6)	52.09±3.87 (42.8-62.5)	51.79±19.8 (22-92)	3.35±0.43 (2.3-4.7)	59.79±4.98 (50.0-80.0)	15.23±5.1 (6.6-27.2)	26.86±2.12 (21.6-34.7)
41-50 n = 1224	1.99±0.2 (1.6-2.6)	50.44±3.91 (40.8-59.3)	38.27±12.7 (20-64)	3.47±0.49 (2.1-5.2)	58.3±4.72 (47.8-69.7)	14.29±4.77 (5.2-25)	27.57±2.54 (21.5-34.2)
51-60 n = 879	2.01±0.26 (1.6-2.7)	51.06±4.44 (40.0-63.8)	37.88±13.5 (16.4-59.2)	3.51±0.54 (2.4-4.6)	57.15±4.16 (50.0-64.8)	14.39±4.75 (5.3-22.5)	28.41±2.82 (22.8-36.7)
61-70 n = 536	2.1±0.22 (1.8-2.7)	49.42±2.98 (43.4-55.5)	37.18±17.1 (15.6-70)	3.50±0.47 (2.5-4.5)	56.79±4.50 (46.0-65.7)	11.76±2.47 (7.4-16.3)	29.39±2.15 (26.2-34.3)
71-80 n = 321	2.27±0.23 (2.0-2.4)	43.17±2.15 (43.7-45.8)	22.91±8.4 (7.63-43.2)	4.63±0.60 (4.0-5.2)	52.4±2.26 (49.9-54.3)	8.99±5.66 (4.8-15.7)	33.27±0.51 (32.7-33.7)

The values are presented as mean±SD, n = number, SL = sensory latency; SNCV = sensory nerve conduction velocity; SNAPA = sensory nerve action potential amplitude, DML = distal motor latency measured from the onset of action potential, MNCV = motor nerve conduction velocity, CMAPA = compound muscle action potential amplitude measured from peak to peak. The values between brackets represent the range.

Table 3: Ulnar Nerve Parameters in Different Age Groups

Age Group (years)	SL (msec)	SNCV (m/sec)	SNAPA (µV)	DML (msec)	MNCV (m/sec)	CMAPA (mV)	F-wave (msec)
< 1 n = 331	1.37±0.15 (1.2-1.5)	35.37±6.65 (30.0-42.8)	38.81±11.4 (16.56-49.7)	2.83±0.4 (2.4-3.2)	46.83±2.78 (44.8-50.0)	10.1±5.52 (6.98-14.3)	16.81±1.13 (14.8-18.1)
1-10 n = 642	1.73±0.29 (1.3-2.6)	48.92±4.50 (40.4-55.8)	49.41±19.85 (19.6-87.3)	2.36±0.47 (1.7-3.3)	55.06±3.89 (46.4-61.3)	12.68±9.8 (5.5-27.2)	21.43±2.6 (14.9-25.1)
11-20 n = 619	1.94±0.18 (1.6-2.5)	54.18±3.80 (44.0-62.5)	58.58±10.11 (48-70.5)	2.53±0.64 (1.8-4.6)	63.07±5.71 (50.0-74.2)	12.61±4.44 (4.88-17.8)	25.69±3.01 (22.7-34.1)
21-30 n = 1213	1.90±0.20 (1.6-2.8)	55.25±3.66 (46.2-64.7)	55.57±24.99 (24-120)	2.30±0.39 (1.6-4.2)	64.27±5.73 (50.0-78.5)	15.95±5.37 (10.3-27.6)	26.29±2.13 (21.9-33.0)
31-40 n = 1419	1.91±0.19 (1.5-2.8)	55.03±3.94 (42.3-68.8)	51.41±16.48 (19.2-110)	2.29±0.34 (1.6-3.9)	64.10±4.91 (50.8-77.6)	13.36±3.68 (5.6-21.8)	25.98±2.41 (21.7-34.9)
41-50 n = 1103	1.95±0.17 (1.6-2.6)	53.76±3.8 (42.0-63.8)	46.89±15.9 (20.2-96)	2.36±0.34 (1.6-3.6)	63.04±5.59 (46.7-78.7)	13.9±5.59 (7.2-31.8)	27.65±2.82 (23.3-35.9)
51-60 n = 766	1.98±0.2 (1.6-2.7)	52.93±3.95 (40.7-62.5)	46.76±16.3 (22.4-83)	2.42±0.34 (1.7-3.7)	61.69±5.35 (50.0-75.6)	13.18±4.57 (6.7-19.6)	27.78±2.73 (23.6-36.7)
61-70 n = 399	2.06±0.2 (1.8-2.7)	50.39±3.44 (44.4-57.8)	34.79±18.12 (8.8-72)	2.56±0.49 (1.8-4.2)	60.48±6.2 (46.8-72.5)	9.65±5.41 (1.9-15.5)	28.79±2.65 (25.7-35.5)
71-80 n = 287	2.26±0.28 (2.0-2.8)	50.7±3.02 (47.5-57.1)	32.17±14.7 (18.6-65.3)	3.0±0.5 (2.3-3.6)	55.74±3.61 (51.6-62.2)	8.65±4.47 (5.94-18.1)	31.88±3.11 (28.8-36.9)
>80 n = 189	3.1±0.28 (2.9-3.3)	49.55±1.63 (48.4-50.7)	27.9±10.4 (11.3-54.1)	3.4±0.71 (2.9-3.9)	53.25±6.15 (48.9-57.6)	7.49±4.85 (3.98-13.6)	33.0±1.75 (30.7-34.7)

The values are presented as mean±SD, n = number, SL = sensory latency; SNCV = sensory nerve conduction velocity; SNAPA = sensory nerve action potential amplitude, DML = distal motor latency measured from the onset of action potential, MNCV = motor nerve conduction velocity, CMAPA = compound muscle action potential amplitude measured from peak to peak. The values between brackets represent the range.

Table 4: Radial Nerve Parameters in Different Age Groups

Age Group (years)	SL (msec)	SNCV (m/sec)	SNAPA (µV)	DML (msec)	MNCV (m/sec)	CMAPA (mV)	F-wave (msec)
< 1 n = 219	1.25±0.07 (1.2-1.3)	49.6±5.94 (45.4-53.8)	36.5±8.43 (18.33-52.3)	1.85±0.07 (1.8-1.9)	51.15±1.63 (50.0-52.3)	8.21±4.32 (5.34-15.6)	19.0±2.12 (17.5-20.5)
1-10 n = 476	1.72±0.26 (1.2-2)	53.41±3.07 (45.4-56.7)	47.22±13.54 (32.4-69.9)	2.2±0.23 (1.8-2.5)	56.62±3.32 (50-60.1)	13.99±10.2 (6.98-20.5)	21.53±1.99 (17.5-24.1)
11-20 n = 559	1.8±0.17 (1.5-2.2)	54.68±4.95 (50.0-68.7)	54.76±11.39 (39.8-74.8)	2.94±0.48 (2.0-4.0)	58.82±4.53 (50.0-69.0)	12.87±7.65 (5.91-19.3)	27.4±2.67 (21.2-30.1)
21-30 n = 984	1.91±0.3 (1.5-2.5)	52.41±4.94 (43.3-62.5)	55.45±17.9 (34.1-95.2)	3.12±0.89 (1.7-5.3)	60.68±7.25 (50.8-77.2)	14.39±9.73 (8.3-21.3)	26.24±3.13 (20.6-31.1)
31-40 n = 1123	1.73±0.24 (1.4-2.3)	54.9±4.41 (47.5-66.6)	50.21±12.1 (24.2-87.9)	2.71±0.66 (1.5-3.4)	61.17±7.36 (53.0-70.0)	13.98±6.77 (8.6-24.8)	28.66±2.9 (23.7-32.2)

41-50 n = 895	1.78±0.31 (1.4-2.4)	53.71±4.4 (45.6-60.7)	45.7±10.3 (28.2-78.9)	3.34±1.01 (1.8-5.0)	54.36±6.79 (47.5-65.9)	13.0±5.11 (6.6-23.8)	29.33±1.9 (25.8-31.3)
51-60 n = 766	2.05±0.5 (1.8-2.8)	53.9±5.59 (49.0-61.1)	44.24±12.83 (18.7-69.9)	3.58±0.56 (2.9-4.4)	58.9±7.92 (52.2-68.5)	13.76±7.57 (5.57-15.6)	28.58±1.04 (27.6-29.9)
61-70 n = 402	2.57±0.45 (2.1-3.0)	50.35±3.14 (47.6-54.8)	37.79±18.2 (18.8-76.3)	3.7±0.47 (3.1-4.2)	54.38±3.7 (50.1-59.1)	10.77±5.29 (2.6-17.3)	29.08±3.02 (25.8-33.1)
71-80 n = 195	2.21±0.31 (2.1-2.9)	48.7±4.52 (44.4-55.4)	27.2±10.96 (17.3-49.73)	3.3±0.54 (2.8-4.2)	50.24±4.16 (46.9-58.4)	9.29±4.88 (3.8-14.4)	33.6±3.12 (29.1-34.9)

The values are presented as mean±SD, n = number, SL = sensory latency; SNCV = sensory nerve conduction velocity; SNAPA = sensory nerve action potential amplitude, DML = distal motor latency measured from the onset of action potential, MNCV = motor nerve conduction velocity, CMAPA = compound muscle action potential amplitude measured from peak to peak. The values between brackets represent the range.

Table 5: Musculocutaneous Nerve Parameters in Different Age Groups

Age Group (years)	**SL (msec)**	**SNCV (m/sec)**	**SNAPA (µV)**	**DML (msec)**	**MNCV (m/sec)**	**CMAPA (mV)**
1-10 n = 211	2.0±0.1 (1.4-2.5)	51.2±2.4 49.2-55.2)	10.1±3.2 (7.9-14.2)	3.98±0.3 (3.6-4.4)	50.4±4.26 (44.7-56.3)	10.9±4.13 (7.3-15.4)
11-20 n = 327	2.3±0.1 (1.8-2.7)	56.3±3.2 50.1-58.2)	12.1±3.2 (8.2-14.2)	4.25±0.64 (3.4-4.9)	59.8±6.33 (52.1-67.6)	13.1±8.64 (7.3-27.4)
21-30 n = 300	2.8±0.3 (2.1-0.5)	65.1±4.5 59.8-69.33)	14.2±5.4 (9.3-16.4)	4.44±0.42 (3.8-5.2)	64.84±6.04 (53.9-71.4)	15.9±7.94 (6.85-26.1)
31-40 n = 380	2.9±0.5 (2.1-0.61)	62.3±4.7 (59.4-69.2)	13.6±4.8 (9.2-17.7)	4.57±0.53 (3.9-5.6)	65.18±6.91 (51.7-76.2)	14.2±6.43 (6.25-24.7)
41-50 n = 433	3.1±0.51 (2.4-0.52)	64.3±5.1 (58.7-68.4)	13.4±5.21 (8.6-16.9)	4.98±0.79 (3.9-6.5)	63.21±6.72 (50.5-73.0)	13.8±5.43 (6.2-22.3)
51-60 n = 265	3.2±0.53 2.6-0.55)	60.3±5.22 (56.9-66.1)	11.4±4.7 (7.3-15.6)	4.71±0.36 (4.0-5.1)	64.69±6.75 (52.1-74.4)	10.6±6.7 (5.7-21.4)
61-70 n = 189	3.3±0.57 (2.7-0.81)	58.3±5.3 (52.8-63.1)	10.3±4.6 (6.2-13.3)	5.33±0.61 (4.8-6.0)	50.33±7.09 (42.7-56.7)	8.65±7.33 (5.7-19.2)

The values are presented as mean±SD, n = number, SL = sensory latency; SNCV = sensory nerve conduction velocity; SNAPA = sensory nerve action potential amplitude, DML = distal motor latency measured from the onset of action potential, MNCV = motor nerve conduction velocity, CMAPA = compound muscle action potential amplitude measured from peak to peak. The values between brackets represent the range.

Table 6: Axillary Nerve Parameters in Different Age Groups

Age Group (years)	**DML (msec)**	**MNCV (m/sec)**	**CMAPA (mV)**
1-10 n = 209	4.56±0.67	46.24±5.24	10.43±4.83
11-20 n = 327	3.58±0.49	54.37±2.41	13.7±5.2
21-30	3.9±0.59	54.91±6.61	17.4±6.87

n = 300			
31-40 n = 376	4.3±0.24	52.87±3.6	11.6±5.1
41-50 n = 420	4.23±0.54	52.3±5.66	12.5±4.8
51-60 n = 265	4.82±0.39	52.18±3.01	9.98±5.73
61-70 n = 180	5.3±0.76	49.88±1.65	8.7±3.7

The values are presented as mean±SD, n = number, DML = distal motor latency measured fro the onset of action potential, MNCV = motor nerve conduction velocity, CMAPA = compound muscle action potential amplitude measured from peak to peak.

Table 7: Tibial Nerve Parameters in Different Age Groups

Age Group (years)	SL (msec)	SNCV (m/sec)	SNAPA (µV)	DML (msec)	MNCV (m/sec)	CMAPA (mV)	F-wave (msec)
< 1 n = 213	2.1±0.29 (1.8-2.5)	35.35±8.52 (24.0-44.7)	12.4±8.23 (7.3-28.9)	2.93±0.94 (2.0-4.2)	45.28±11.3 (35.0-57.5)	7.5±4.6 (4.1-15.4)	24.76±2.61 (21.0-28.2)
1-10 n = 422	2.56±0.56 (1.5-4.2)	46.48±5.48 (30.7-57.1)	15.2±10.5 (8-32.3)	3.02±0.73 (1.8-4.7)	50.46±5.45 (40.7-64.0)	10±6.65 (5.1-18.2)	30.15±5.84 (21.7-45.2)
11-20 n = 513	4.0±0.55 (2.4-4.7)	46.18±4.22 (40.0-58.7)	15.6±6.57 (5.2-21.9)	3.82±0.5 (2.4-4.7)	51.49±4.73 (42.3-65.5)	9.8±4.78 (4.2-19.6)	46.9±5.06 (38.3-56.8)
21-30 n = 822	4.23±0.52 (3.0-5.7)	44.19±3.89 (36.0-57.8)	13.26±5.28 (9.2-28)	4.08±0.65 (2.9-6.3)	50.29±5.67 (41.7-66.9)	8.7±4.53 (3.2-16.4)	48.74±4.45 (40.4-57.5)
31-40 n = 1213	4.17±0.53 (3.0-5.5)	43.93±4.28 (33.0-55.0)	10.97±3.9 (4.8-16.2)	4.08±0.61 (3.0-5.5)	50.6±5.3 (40.2-65.9)	9.95±3.5 (5.1-15.7)	49.02±4.34 (40.0-61.0)
41-50 n = 1544	4.34±0.51 (3.2-5.9)	42.68±3.68 (33.0-54.4)	9.95±3.39 (7.7-20.8)	4.25±0.62 (3.0-5.9)	48.92±5.05 (40.0-61.8)	8.5±4.93 (4.2-18.5)	51.45±4.39 (41.5-61.8)
51-60 n = 877	4.27±0.42 (3.3-5.0)	41.39±4.71 (24.1-50.0)	9.98±4.6 (6.8-18.6)	4.01±0.61 (3.0-5.5)	47.24±4.0 (41.8-57.1)	9.54±5.34 (4.3-16.9)	51.55±5.12 (44.6-64.1)
61-70 n = 419	4.37±0.57 (3.5-5.5)	40.64±4.08 (32.6-47.3)	8.95±7.1 (6-15.9)	4.35±0.72 (3.3-6.9)	46.65±4.41 (38.3-57.0)	6.2±3.28 (3.8-10.4)	52.36±5.53 (42.5-61.9)
71-85 n = 185	4.89±0.74 (4.0-6.2)	37.54±3.63 (32.2-41.8)	5.67±8.4 (3.5-16.7)	4.64±1.3 (2.5-8.4)	46.71±3.59 (38.5-51.6)	5.9±4.1 (4.1-11.5)	53.48±4.74 (49.3-63.2)

The values are presented as mean±SD, n = number, SL = sensory latency; SNCV = sensory nerve conduction velocity; SNAPA = sensory nerve action potential amplitude, DML = distal motor latency measured from the onset of action potential, MNCV = motor nerve conduction velocity, CMAPA = compound muscle action potential amplitude measured from peak to peak. The values between brackets represent the range.

Table 8: Common peroneal Nerve Parameters in Different Age Groups

Age Group (years)	SL (msec)	SNCV (m/sec)	SNAPA (µV)	DML (msec)	MNCV (m/sec)	CMAPA (mV)	F-wave (msec)
< 1 n = 209	2.07±0.28 (1.8-2.6)	33.93±5.87 (25.8-42.1)	8.99±7.61 (4.1-15.7)	2.63±0.67 (1.8-3.9)	47.88±5.46 (40.0-54.0)	4.3±3.11 (2.6-7.91)	26.13±2.92 (24.4-29.5)
1-10 n = 420	2.48±0.43 (1.9-3.5)	41.92±4.25 (31.0-50.0)	14.47±8.11 (9.8-24.6)	2.77±0.61 (1.9-4.3)	48.27±3.6 (43.0-55.0)	9.11±4.1 (4.5-12.4)	31.46±7.08 (24.2-45.3)
11-20 n = 513	3.31±0.44 (2.0-4.0)	43.42±3.25 (38.2-52.5)	15.6±6.57 (7.85-26.4)	3.77±0.51 (2.5-4.7)	50.48±4.1 (43.8-60.6)	9.2±5.12 (5.7-14.8)	47.65±3.21 (41.6-51.2)
21-30 n = 822	3.51±0.39 (2.6-4.3)	43.39±3.33 (37.8-55.1)	11.67±5.4 (5.8-21.2)	3.64±0.62 (2.4-5.5)	50.6±5.07 (40.0-68.0)	8.9±2.31 (5.6-10.9)	47.68±3.86 (40.5-58.4)
31-40 n = 1213	3.42±0.45 (2.2-4.5)	43.5±3.88 (33.3-63.6)	13.2±7.52 (4.8-23.7)	3.75±0.62 (2.2-5.4)	51.11±4.34 (42.7-60.0)	8.13±4.29 (4.8-13.3)	48.71±4.23 (40.4-59.5)
41-50 n = 1534	3.62±0.36 (3.2-4.6)	40.87±3.16 (31.5-45.7)	14.8±10.58 (7.6-28.2)	3.82±0.73 (2.7-5.7)	49.2±4.19 (40.1-58.0)	9.72±4.02 (4.1-13.8)	48.91±5.09 (41.1-59.9)
51-60 n = 860	3.56±0.42 (3.0-4.8)	40.22±3.91 (32.0-48.4)	10.15±5.35 (4.9-17.4)	3.8±0.71 (2.7-5.2)	47.32±4.51 (39.5-55.2)	8.43±5.1 (3.8-14.7)	50.24±4.26 (42.9-57.7)
61-70 n = 400	3.5±0.29 (3.1-4.1)	40.59±2.36 (36.7-45.1)	9.9±6.8 (3.9-16.8)	3.59±0.42 (2.9-4.2)	48.19±4.53 (41.1-60.0)	5.58±3.31 (2.7-11.8)	49.92±4.53 (40.8-56.1)
71-85 n = 180	3.83±0.63 (2.7-4.7)	38.81±3.68 (32.6-44.4)	6.41± 3.5 (3.1-10.9)	3.51±0.46 (3.0-4.5)	48.53±3.66 (44.1-55.7)	5.86±2.76 (3.2-10.8)	50.49±2.66 (47.0-54.0)

The values are presented as mean±SD, n = number, SL = sensory latency; SNCV = sensory nerve conduction velocity; SNAPA = sensory nerve action potential amplitude, DML = distal motor latency measured from the onset of action potential, MNCV = motor nerve conduction velocity, CMAPA = compound muscle action potential amplitude measured from peak to peak. The values between brackets represent the range.

Table 9: Femoral Nerve Parameters in Different Age Groups

Age Group (years)	DML (msec)	MNCV (m/sec)	CMAPA (mV)
1-10 (n = 155)	3.1±0.12	50.1±4.7	9.11±4.1
11-20 (n = 344)	3.68±0.18	55.42±5.3	9.2±5.12
21-30 (n = 426)	4.38±0.6	53.5±5.03	8.9±2.31
31-40 (n = 587)	4.53±0.4	50.03±8.11	8.13±4.29
41-50 (n = 630)	5.07±0.23	52.7±4.37	9.72±4.02
51-60 (n = 288)	5.33±0.46	46.8±3.31	8.43±5.1
61-70 (n = 276)	6.0±0.96	41.43±4.2	5.58±3.31

The values are presented as mean±SD, n = number, DML = distal motor latency measured from the onset of action potential, MNCV = motor nerve conduction velocity, CMAPA = compound muscle action potential amplitude measured from peak to peak.

Table 10: Comparison of nerve conduction parameters of facial, musculocutaneous, axillary, and femoral nerves between the present study and those reported by others.

Facial Nerve	Present study (n = 1793)	Kimura[19]	Waylonis & Johnson[20] (n = 78)	Taylor et al[21]	
DML	3.14±0.37	2.9±0.4	3.4±0.8	4.0±0.5	
MCV	31.07±2.74	----	----	----	
CMAPA	8.75±3.61	----	----	----	
Axillary Nerve	Present study (n = 1868)	Kraft[22] (n = 62)			
DML	4.31±0.73	3.9±0.5			
MCV	52.98±4.34	----			
CMAPA	10.9±3.62	----			
Musculocutaneous Nerve	Present study (n = 1894)	Kraft[22] (n = 62)	Izzo et al[23] (n = 155)	Reddy et al[24] (n = 30)	Spindler & Felsenthal[25] (n = 30)
SL	2.8±0.3	4.5±0.6	2.7±0.2	2.7±0.2	1.8±0.1
SNCV	65.1±4.5	----	63.0±5.0	66.0±4.0	65.0±3.6
SNAPA	14.2±5.4	----	11.4±5.2	15.4±4.1	24.0±7..2
DML	4.68±0.62	----	----	----	----
MCV	62.25±7.92	----	----	----	----
CMAPA	15.05±4.2	----	----	----	----
Femoral Nerve	Present study (n = 2551)	Uludag et al[15] (n = 16)	Gassel [26] (n = 42)	Johnson et al[27]	
DML	4.51±0.71	4.6±0.5	3.7±0.45	6.0±0.7	
MCV	52.07±5.56	----	70.0±5.5	69.4±9.2	
CMAPA	8.44±3.16	----	----		

The values are presented as mean±SD, n = number, SL = sensory latency, SNCV = sensory nerve conduction velocity, SNAPA = sensory nerve action potential amplitude, DML = distal motor latency, MNCV = motor nerve conduction velocity, CMAPA = compound muscle action potential amplitude

Table 11: Comparison of nerve conduction parameters of median, ulnar, and radial nerves between the present study and those reported by others.

Median Nerve	Present study (n = 5766)	Hennessey et al[8] (n = 44)	Shehab[14] (n = 50)	Awang et al[16] (n = 250)	Kimura[18] (n = 65)	Karagoz et al[28] (n = 17)
SL	1.93±0.22	2.5±0.2	2.3±0.3	----	2.84±0.34	----
SNCV	51.9±4.04	61.2±4.3	56.6±7.6	54.04±7.02	56.2±5.8	61.4±3.0
SNAPA	49.5±23.1	31.4±8.2	71.3±23.9	----	38.5±15.6	36.6±6.6
DML	3.39±0.47	3.2±0.4	3.1±0.3	----	3.49±0.34	----
MNCV	58.97±4.8	59.5±4.4	56.5±3.5	54.71±5.69	57.7±4.9	59.3±3.2
CMAPA	14.8±4.92	12.1±3.8	11.1±2.8	----	7.0±3.0	8.3±1.4
Ulnar Nerve	Present study (n = 5519)	Shehab[14] (n = 50)	Awang et al[16] (n = 250)	Kimura[18] (n = 65)	Karagoz et al[28] (n = 17)	Buschbacher[29] (n = 248)
SL	1.94±0.19	2.0±0.2	2.0±0.2	2.54±0.29	----	----
SNCV	54.2±3.97	52.1±7.5	52.1±7.5	54.8±5.3	58.5±3.7	----
SNAPA	50.1±19.8	59.2±17.6	59.2±17.6	35.0±13.4	29.7±4.0	----
DML	2.35±0.39	2.4±0.3	2.4±0.3	2.59±0.39	----	3.0±0.3
MNCV	63.2±5.61	60.4±5.2	60.4±5.2	58.7±5.1	59.7±3.7	61.0
CMAPA	13.8±4.86	9.2±2.2	9.2±2.2	5.7±2.0	12.3±2.0	11.6±2.1
Radial Nerve	Present study (n = 4729)	Falco et al[9] (n = 51)	Johnson et al[13] (n = 78)	Shehab[14] (n = 50)	Jebsen[30] (n = 49)	Trojaborg & Sinrup[31]
SL	1.94±0.33	2.0±0.2	2.4±0.2	1.95±0.3	----	----
SNCV	53.44±4.42	----	----	53.1±8.7	----	58.0±6.0
SNAPA	26.9±13.98	16.5±13.8	12.0±1.0	18.6±5.5	----	13.0±7.5
DML	3.27±0.72	----	----	----	----	----
MNCV	58.84±6.32	----	----	----	61.1±5.9	62.0±5.1
CMAPA	16.1±5.23	----	----	----	----	14.0±8.8

The values are presented as mean±SD, n = number, SL = sensory latency, SNCV = sensory nerve conduction velocity, SNAPA = sensory nerve action potential amplitude, DML = distal motor latency, MNCV = motor nerve conduction velocity, CMAPA = compound muscle action potential amplitude

Table 12: Comparison of nerve conduction parameters of tibial and common peroneal nerves between the present study and those reported by others.

Tibial Nerve	Present study (n = 5388)	Kimura[18] (n = 59)	Buschbacher[32] (n = 250)	Budak[33] (n = 30)	Antunes et al[34] (n = 51)
SL	4.22±0.52	----	----	----	4.84±0.71
SNCV	43.45±4.35	----	----	----	46.1±5.68
SNAPA	12.73±5.2	----	----	----	2.13±1.78
DML	4.09±0.63	3.96±1,0	4.5±0.8	----	4.36±0.67
MNCV	49.59±5.26	48.5±3.6	44-51	47.1±3.0	----
CMAPA	8.97±4.45	5.8±1.9	12.7±4.4	----	20.25±7.57
Common Peroneal Nerve	Present study (n = 5342)	Kimura[18] (n = 60)	Karagoz et al[28] (n = 17)	DiBenedetto[35] (n = 50)	Buschbacher[36] (n =242)
SL	3.49±0.41	----	----	2.24±0.49	----
SNCV	42.5±3.71	----	----	47.3±3.4	----
SNAPA	14.26±7.86	----	----	13.9±4.0	----
DML	3.73±0.63	3.77±0.86	----	----	4.8±0.8
MNCV	49.96±4.76	48.3±3.9	57.4±3.6	----	46.5±4.5
CMAPA	8.22±3.61	5.1±2.3	7.6±1.6	----	6.8±2.5 (young) 5.1±2.5 (old)

The values are presented as mean±SD, n = number, SL = sensory latency, SNCV = sensory nerve conduction velocity, SNAPA = sensory nerve action potential amplitude, DML = distal motor latency, MNCV = motor nerve conduction velocity, CMAPA = compound muscle action potential amplitude

Table 13: Comparison of F-wave latency of median, ulnar, radial, tibial, and common peroneal nerves between the present study and those reported by others.

Nerve	Present study	Alavian-Ghavanini & Haghpanah[17] (n = 73)	Kimura[18] (n = 61)	Budak[33] (n = 30)	Buschbacher[37-39]
Median (n = 3244)	27.23±2.46	-----	26.6±2.2	-----	26.8±2.4 (n = 195)
Ulnar (n = 4681)	26.86±2.41	-----	27.6±2.2	25.7±2.6	-----
Radial (n = 2199)	27.74±2.78	-----	-----	-----	-----
Tibial (n = 2877)	49.74±4.87	46.54±3.94	47.7±5.0	48.0±3.9	50.8±5.3 (n = 159)
Common peroneal (n = 2084)	48.59±4.26	46.06±3.85	48.4±4.0	-----	50.2±5.5 (n = 180)

The values are presented as mean±SD, n= number

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Endoscopic Sinus Surgery versus Conventional Method in Management of Naso-Ethmoidal polyps and Their Associated Intranasal Abnormalities

Hiwa As'ad Rawandzi *FICMS; CABS (ENT*).

Abstract

Background: This is a prospective and comparative clinical study, implemented in department of Otolaryngology –Sulaimani Teaching Hospital, from Aug. 1st 2006 to Nov. 1st 2007.

Objectives: This study was carried out to compare the influences and outcomes of endoscopic sinus surgery versus conventional intranasal method in management of patients with nasal polyps, which is the most common intranasal swelling.

Methods: The sample of the study includes 50 patients' aged 12-75 yeas old that are managed for nasal polyp, thirty patients managed by conventional method and twenty patients were managed by endoscopic sinus surgery.Patients are observed postoperatively by symptomatic score and endoscopically

Results: Endoscopic sinus surgery resulted in better improvement in symptoms,better treatment of other associated sinonasal pathologies, less complication rate,and less recurrence rate than conventional polyoectomy.On the other hand endoscopic sinus surgery is more technically demanding and needs more operative time than the conventional way.

Conclusion: We concluded from the study that Endoscopic Sinus surgery is better than conventional intranasal polypectomy, as endoscopy provides approximate field of vision and illumination, good access, hidden pathology are revealed and managed, and complication, recurrent rate are less.

Keywords: nasal polyp, conventional polypectomy, endoscopic sinus surgery.

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Introduction

The introduction of endoscopes in sinus surgery has brought about a revolution in the approach to surgery of paranasal sinuses. This technical achievement has been critical in the evolution of a functional philosophy of sinus surgery which was introduced by Messerklinger (1978) and Wigand (1981) and further refined by Stammberger (1986) [(1)].

Endoscopic sinus surgery is a minimally invasive technique in which sinus air cells and sinus ostia are opened under direct visualization [(2)].

Teaching Hospital, College of Medicine, University of Sulaimani.
Adress Correspondence to: Dr. Hiwa As'ad Abdulkareem.
College of Medicine/University of Sulaimaniyah.
E.mail: hiwa_ent@yahoo.com
Cell phone: 07701580087
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Most rhinologists agree that Endoscopic Sinus Surgery should be a "disease-directed" and mucosal-sparing operation, recognizing the principle of the potential for re-establishing drainage and mucosal recovery of the dependent sinuses [(3)].

The ability to treat paranasal sinus disease and the nasal cavity has been revolutionized by endoscopes and computed tomographic scanning (CT). Endoscopes have made it possible to examine the nose thoroughly from the anterior nares to the postnasal space [(2)].

Endoscopy also used for therapeutic purposes for patients who are correctly documented disease have failed to respond to appropriate medical therapy like chronic rhinosinusitis and nasal polyp with preservation of mucosal and bony framework in critical areas of the nasal cavity [(4)].

CT scanning identifies the anatomic relationships of the key structures (orbital contents, optic nerve and carotid artery) to the diseased areas. A process that is vital for surgical planning. CT also defines the extent of disease in any individual sinus, as well as any underlying anatomic abnormalities that may predispose a patient to sinusitis and nasal polyp [(2)].

The reasons and concepts supporting the use of FESS have recently become widely accepted. The term "functional" was introduced to distinguish this type of endoscopic surgery from nonendoscopic, "conventional" procedures; the goal of FESS is to return the mucociliary drainage of the sinuses to normal function. The paranasal sinuses are maintained in a healthy state by ventilation through the individual ostia and by a mucociliary transport mechanism that keeps a continuous protective layer of mucus flowing out of the sinuses [1, 2, and 5].

Nasal Polyps are parts of an inflammatory reaction involving the mucosa of the nose and paranasal sinuses. Nasal polyps are evaginations of the nasal mucosa attached by a pedicle arising from the ethmoidal sinuses, middle turbinate, maxillary sinuses and some times from the septum [6].

Types:

- Simple.
- Neoplastic.
  - Benign.
  - Malignant.

There are 5 main theories of pathogenesis:

1. Bernoulli phenomenon.
2. Polysaccharide changes.
3. Vasomotor imbalance.
4. Infection.
5. Allergy [8].

No single predisposing disease can be implicated in the formation of polyps, though they may be associated with several other diseases, notably non-allergic (intrinsic) asthma and aspirin intolerance or sensitivity [9].

The association of asthma and nasal polyps has been noted by numerous authors. What is also recognized, but much less often cited, is that this association is strongest in women [10].

In advanced cases the external nose may be broadened, the condition is known as "frog-face"

On examining the interior of the nose, attention should first be directed to the region of the middle turbinate, though in obvious cases a view of this part will be obscured by the multiple grey polypoidal masses. They are insensitive to probing, mobile on their pedicles, and may totally fill the nose bilaterally.

Posterior rhinoscopy should also be done; the nose can be looked perfectly normal anteriorly in the presence of a large choanal polyp, the narrow pedicle of which cannot be seen in the middle meatus [12].

- There is no specific hematological, biochemical or immunological investigations that are required apart from those involved in the general work up of cases prior to surgery.
- Radiology

Plain radiographs of the paranasal sinuses demonstrated by the conventional three views will show the extent of the disease in the nose and paranasal sinuses to some extent. A CT_ scan will give more information, particularly the anatomical detail [8].

Management:

The treatment of nasal polyposis is a debatable subject. Surgical or medical treatment or both have been recommended as the treatment of choice.

A.Medical treatment:

According to the "Position statement on nasal polyps," medical treatment should be used for at least 1 month before surgery is contemplated in patients with typical nasal polyposis because some studies have indicated that in those patients who respond to medical treatment , no additional treatment is necessary[13].

Steroids are the cornerstone for treating NP. This can range from topical steroid sprays or drops in mild to moderate polyposis, to a short course of systemic steroids in severely affected patients[14].

Both oral corticosteroids and topical nasal corticosteroids are effective in shrinking polyps and controlling their

recurrence. Topical corticosteroids are first-line therapy that should be employed prior to considering surgical intervention. Unless there is a contraindication, a trial of a tapering course of oral corticosteroids is also frequently used prior to surgical resection. Should surgery eventually become necessary, topical corticosteroids and occasionally oral corticosteroids may be needed for long-term maintenance [(15)].

B.Surgical treatment:

Anaesthesia

Surgical treatment is performed under either local or general anesthesia. If the operation is performed under local anesthesia, the polyps themselves often shrink, which makes removal difficult. However, recurrence is common as there are large parts of the polyps which shrink into the ethmoidal cells.

General anesthesia on the other hand allows the surgeon an excellent access to the ethmoidal polyps [(16)].

Conventional polypectomy

A snare is passed around each polyp in turn and gradually tightened as high up around the pedicle as possible .It is important that the pedicle should be avulsed and not cut or torn through, otherwise early recurrence is inevitable [(12)].Or forceps are used to remove the polyps.

Endoscopic sinus surgery

Polyps are removed using endoscopy;and associated pathologies in the nose and paranasal sinuses are corrected through endoscopic sinus surgery.

Patients and the method:

The present prospective comparative study was conducted in the department of otolaryngology, endoscopy unit, Sulaimani Teaching Hospital from August 2006 to November 2007.

The sample of the study consists of 50 patients aged above 12 years old who underwent nasal polypectomy either conventionally (30 patients) or endoscopically(20 patients). All were otherwise healthy, and all provided informed consent.

Patients having antrochoanal polyp, marked deviation of nasal septum and nasal or antral tumour were excluded from the study.

A. Initial patient work-up: included detailed history taking, the symptoms and their duration. Thereafter, detailed examination including anterior rhinoscopy, posterior rhinoscopy, and endoscopical examination, and throat, ear examination was done. These procedures are done through rigid nasal endoscope; Hopkins rod which had diagnostic and therapeutic applications; the great and single advancement brought about by endoscopy is its ability to assist in the accurate diagnosis of sinonasal diseases for this 4mm zero or 30° Hopkins rod are used after anesthetizing the area by 4% xylocaine spray. Also using nasal endoscopy provides excellent illumination through endoscope, good acsess and better vi

sualization without magnification that can be transmitted by optical camera to a monitor.

The whole nasal cavity can be visualized in approximate view by three passes of endoscope along the floor of nasal cavity, between middle and inferior turbinate and between the middle turbinate and septum of the nose. Each area of nasal cavity is examined in a systematic fashion [4, 5]. . The specific features that must be identified and assessed during the examination are the middle turbinate and the middle meatus (osteomeatal complex), anatomic obstruction like septal deviation near ostiomeatal complex and the presence of mucopus and nasal polyps [(4)].

All patients were given medical treatment for two weeks in the form of broad-spectrum antibiotics, antihistaminics and local or systemic steroids and decongestants. The patients were then subjected to compute tomography scan of

paranasal sinuses- both axial and coronal views.

B.Definate treatment:The patients are divided into two groups:.

1.Twenty patients underwent endoscopic sinus surgery for nasal polyps.

The extent of surgery was decided based on the findings in pre-operative endoscopy and CT scan of paranasal sinuses. Uncinectomy, removal of lateral wall of concha bullosa, anterior ethmoidectomy, posterior ethmoidectomy, middle meatus antrostomy and/or clearance of frontal recess were performed in the second group of patients. Sphenoid sinus ostium was widened only if CT scan showed evidence of its involvement. Along with this any significant anatomical abnormality was also noted and taken in consideration during surgery.

2.Thirty patients underwent conventional intranasal polypectomy.A snare is passed around each polyp in turn and gradually tightened as high up around the pedicle and avulsed;or forceps are used to remove polyps.

C. Post operative follow up: At the time of discharge from the hospital, the patients were given systemic antibiotic for 10 days along with decongestant drops. Steroid nasal spray was advised in all cases. Alkaline nasal douching was also advised. Patients were advised follow-up one week later, six weeks and three months. Subjective assessment for symptomatic improvement was done and objective results were assessed by check endoscopy. The results were then compiled.

Statistical analysis

The data are analysed by using StatCalc, version 5.3.4, by chi square analysis, P value less than 0.05 regarded as significant.

Results

The first operative group is composed of 13 males and 7 females and the second operative group consists of 20 males and 10 females.

The mean age of first operative group is 34.3 years while the mean age of second operative group is 37.9 years. There is no significant difference between the two groups regarding the age and sex, p value 0.38 (see table 1).

Table 1: patient's characteristics Distribution of number, age and sex between the two groups.

characteristics	group I	group II
Number of patients	20	30
Mean age (years) ± SD*	34.3±13.09	37.9±14.78
Sex (M: F) ratio	1.85:1	2:1
Residence (urban /Rural)ratio	1.5:1	1.5:1

Group I: endoscopic sinus surgery group.
Group II: conventional intranasal polypectomy group.
SD = Standard deviation
* = p value = 0.38

The occupations of the patients are shown in the (Figure 1).

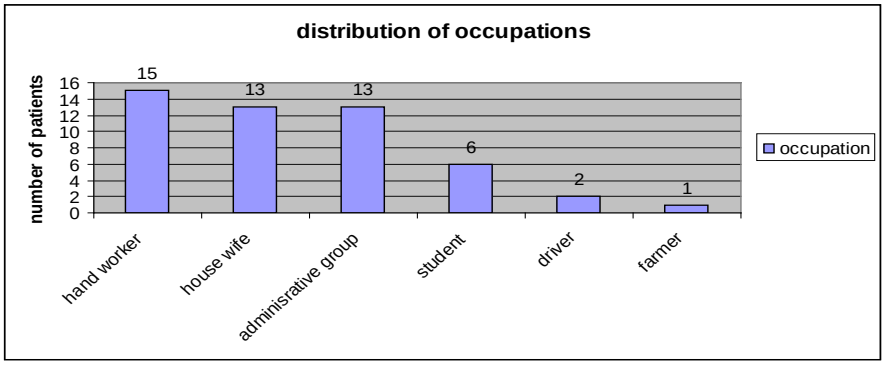

Figure 1: shows the occupation distribution in the study group

Regarding the preoperative symptoms, both groups had nearly similar complaints, all they had nasal polyps with exclusion of antrochoanal polyp, nasal and antral tumor.

Table 2: preoperative symptoms.

Symptoms	**group I n=20**	**group II n=30**
1-nasal obstruction	20 (100%)	22 (73.3%)
2-running and sneezing	18 (90%)	29 (96.6%)
3- hyposmea	20 (100)	22(73.3%)
4-pain	18 (90%)	29(96.6%)
5-postnasal drip	5 (25%)	8 (26.6%)
6-hyponasal speech	20 (100%)	26 (86.6%)
7-mouth breathing	16 (80%)	18 (60%)

n= number

Regarding the preoperative examination by anterior/posterior rhinoscopy and nasal endoscope as a diagnostic tool for the two groups, gross finding such as deviated nasal septum, and hypertrophied turbinate are noted by either techniques while a hidden pathologies such as tiny middle meatal polypi and deformities of the middle turbinate are clearly visible by nasal endoscope (Figure2).

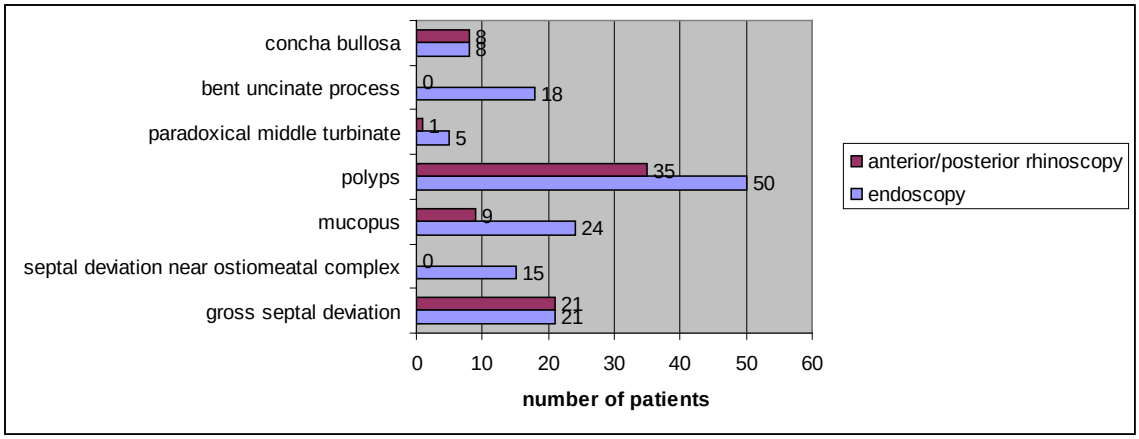

Figure 2: comparison of nasal pathology on clinical examination.

Regarding the operative procedures, Group I underwent Endoscopic sinus surgery for management of nasal polyps and associated finding preoperatively and intraoperativly while all group II patients underwent conventional intranasal polypectomy (table 3).

Table 3: management of associated intranasal abnormality by Endoscopic Sinus Surgery.

Surgical Procedures	**number n=20**	**%**
Septoplasty	10	50%
Uncinectomy	18	90%
Anterior /posterior ethmoidectomy	18	90%
Removal of lateral wall of choncha bullosa	8	40%
Agger nasi exenteration	6	30%
Enlargement of maxillary sinus ostium	14	70%
Frontal recess clearance	1	50%

n=normal.

The duration of operation lasted longer in group I, the operation for 16 patients lasted 1.5-2 hours whereas 4 patients' lasted 1-1.5 hours. While the duration for operation in group II lasteed shorter, the operation for 25 patients' lasted 0.5-1 hours, 4 patients lasted less than 0.5 hour and only one patient last 1-1.5 hours (figure 3), p value (0.000).

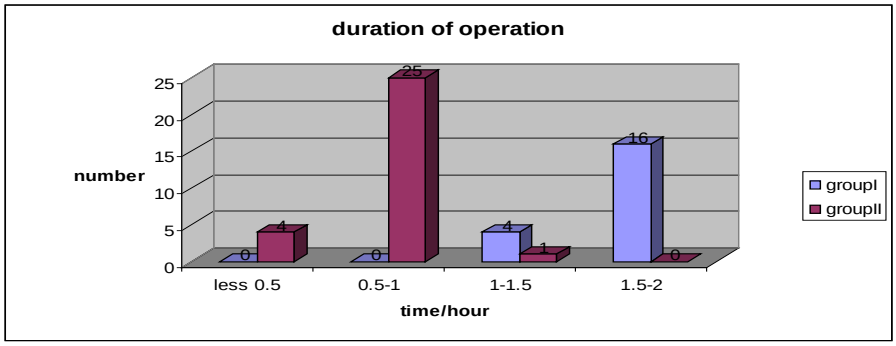

Figure 3: Duration of operation.

During follow up to six months postoperatively, patients in group I show great improvement symptomatically in comparison with symptomatic improvement in group 2, see table (4).

Table 4: postoperative finding

Symptom	Group I n=20	Group II n=30	p value
nasal obstruction	2 (10%)	13 (43.3%)	0.001
running and sneezing	4 (20%)	15 (50%)	0.03
hyposmea	2 (10%)	13 (43.3%)	0.001
pain	2 (10%)	10 (33.3%)	0.07
postnasal	4(20%)	12 (40%)	0.13
hyponasal speech	10 (0.0%)	2 (10%)	0.23
mouth breathing	10 (0.0%)	2 (10%)	0.23

n=number

From 20 patients, only 2 patients remain with nasal obstruction postoperatively in group I which represent 10% of patients (see figure 4). While from 30 patients, 8 patients remain with nasal obstruction which represents 26.6% of patients (see figure 5).

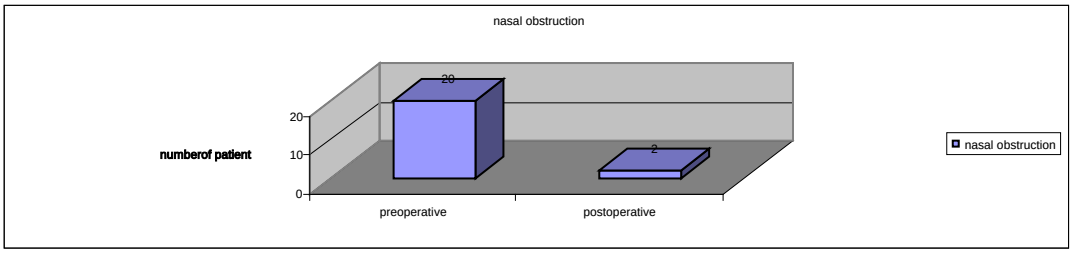

Figure 4: Summary of preoperative and post operative nasal obstruction in group I.

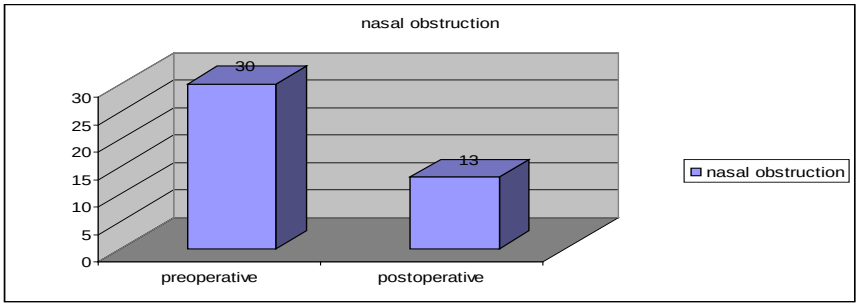

Figure 5: summary of preoperative and postoperative nasal obstruction in group II.

From 20 patients, only 2 patients remain with hyposmea postoperatively in group I which represent 10% of patients ;(Figure6). While from 22 patients, 13 patients remain with hyposmea postoperatively in group II which represent 59% of patients; (Figure 7).

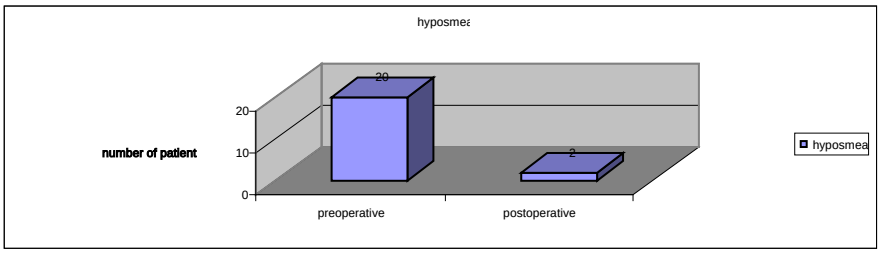

Figure 6: summary of preoperative and postoperative hyposmea in group I.

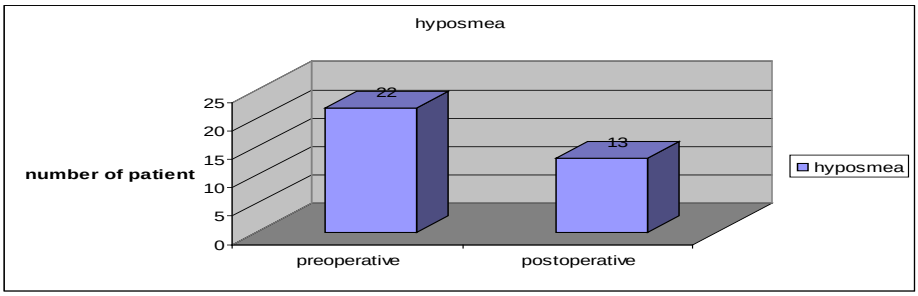

Figure 7: summary of preoperative and postoperative hyposmea in group II.

Regarding complications, minor complications like crusting present in 60% of patients in group I while 26.6% complain of crusting postoperatively.

Because of associated pathology that had been corrected in group I; one patient developed septal haematoma (Table5).

Table 5: complications of operations

Complications	Group I	Group II	P value
Intranasal Crusting	12(60%)	8(26.6%)	0.018
Epistaxis	3(15%)	7(23.3%)	0.47
Septal hematoma	1(5%)	0	0.21
Periorbital chymosis	1(5%)	1(3.3%)	0.76
Rhinitis/sinusitis	1(5%)	5(16.6%)	0.21
Surgical emphysema	0	0	
Orbital cellulitis	0	0	
Orbital hematoma	0	0	
Meningitis	0	0	

During follow up postoperatively, recurrence of polyps in group I found in only 1 patient which represent (5%) of the patients, while the recurrence group II found in 9 patients which represents (30%) of the patients.

In group I, syncline found in only two patients (10%), while in group II, it is found in 5 patients (16.6%) during follow up examination. (Table6)

Table 6: Endoscopic finding on follow up.

Finding	Group I n=20	Group II n=30	P value
Recurrence of polyps	1 (5%)	9(30%)	0.03
Synechiae	2(10%)	5 (16.6%)	0.33

Discussion

The results of 50 patients with ethmoidal polyps undergoing Endoscopic Sinus Surgery (ESS) or conventional intranasal polypectomy are analyzed.

Thirty patients are undergoing conventional intranasal polypectomy and twenty patients undergoing endoscopic sinus surgery.

There are no significant differences between the two groups regarding the age and sex.The urban/rural ratios are the same between the two groups P value (0.38), (see table1).

Regarding the preoperative symptoms, both groups have nearly similar complaints. They have nasal polyps with exclusion of antrochoanal polyp, nasal and antral tumor.

Concerning the preoperative examination by anterior/posterior rhinoscopy and nasal endoscope as a diagnostic tool for the two groups, gross finding such as deviated nasal septum, and hypertrophied turbinate are noted by either techniques while a hidden pathologies such as tiny middle meatal polypi and deformities of the ostiomeatal complex are clearly visible by nasal endoscope (Figure2). This concludes that nasal endoscopies is much more superior to

anterior/posterior rhinoscopy to examin and diagnose intranasal pathologies. This coincide with the study of MATTOX, D.E [17] and LEVINE,H.L[18] who stressed on the importance of endoscopy in the diagnosis of sinonasal diseases .

The diagnosis of recurrent ethmoiditis due to a small polyp obstructing the ostiomeatal complex can be difficult and often unrecognized during anterior rhinoscope examination and can be diagnosed by nasal endoscope [19].

Regarding the operative procedures , all patients in group II underwent just conventional intranasal polypectomy while apart from polypectomy other pathologies were treated in group I like septoplasty (50%) , uncinectomy (90%), anterior/posterior ethmoidectomy (90%), removal of lateral wall of concha bullosa (40%), Agger nasi exenteration(30%), Enlargement of maxillary sinus ostium(70%) and Frontal recess clearance(5%), (Table 3).This may explain that with endoscopic surgeries we can treat other pathologies which can not be treated by conventional way.

This validates the result of Ms Kim Dalziel, Research Fellow (LEAD), who states that ESS aims not only to remove the polyp but also to improve sinus drainage and ventilation, which may decrease recurrence rates. Advantages are claimed over conventional surgery: permitting a better view of the surgical field, fewer complications and lower recurrence rates. ENT specialists, aside from removal of polyps, use endoscopes for a variety of procedures [20].

The duration of operation lasts longer in group I, the operation for 16 patients lasts 1.5-2 hours whereas 4 patients' last 1-1.5 hours. While the duration for operation in group II lasts shorter, the operation for 25 patients' lasts 0.5-1 hours, 4 patients lasts less than 0.5 hour and only one patient lasts 1-1.5 hours (Figure 3), p value (0.000).

The final follow up was done after six months of surgery. There is satisfactory relief of symptoms postoperatively in all the subjects of the study with significant difference regarding nasal obstruction,running and sneezing and hyposmia (p value 0.001,0.03.0.001 respectively). These could be attributed to the removal of polyps and partly to the practice of doing anterior/ posterior ethoidectomy and other associated procedures of the middle turbinate, which bears the brunt of inspiratory airflow (See table 4). Stammberger (1991) reports that 85% very good overall improvement and facial pain and headache were assessed as better as 93.4% [21].

The most common complication encountered involving intranasal crustation in 12 patients (60%), epistaxis in 3 patients (15 %) and intranasal adhesions in 2 patients (10%).

Close outpatient postoperative care with meticulous cleaning of the nasal cavity under endoscopic guidance can easily prevent most of these adhesions.

The patients are advised to use nasal saline irrigation two to three time's daily and nasal ointment to prevent dryness and crusting for at least two months postoperatively. The use of gel film has been reported to be effective in preventing synechiae formation between the middle turbinate and the lateral nasal wall.

The recurrence rate after six month period of follow up shows significant differences in recurrence rate , in group II, 9 patients (30%) while in group I shows only 1 patient (5%) , p value 0.03.

We strongly recommend using edoscopic examination even in those departments which do not routinely

perform endoscopic sinus surgery, as it often reveals hidden pathologies.

Endoscopic sinus surgery is better than conventional intranasal polypectomy because of other pathologies can be treated like high septal deviation, tiny polyp obstructing the ostiomeatal complex and better ventilation and drainage of nose and paranasal sinuses.

The recurrence rate and complication are less if compared with conventional intranasal polypectomy.

It's a minimally invasive surgery, has approximate field of vision and illumination and good acsess if compared with conventional intranasal polypoectomy, but it needs well trained and expert surgeon.

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Pulmonary Hypertension in Patients with Chronic Renal Failure

Jawad Kadhem Manuti *FICMS* .

Abstract

Background: Pulmonary hypertension (PH) comprises a group of clinical and pathophysiological entities with similar features but a variety of underlying causes. Many etiologies causing PH have been reported, and one of the background disease seen with patients with PH is chronic renal failure (CRF).

Objective: To evaluate the prevalence of pulmonary hypertension in patient with chronic renal failure in predialysis state and in uremic patients undergoing hemodialysis

Patients and methods: One hundred patients had complaining chronic renal failure. 50 patients on conservative treatment and 50 patients on hemodialysis evaluated clinically and by echocardiography for the presence of pulmonary hypertension

Results: The prevalence of pulmonary hypertension >35 mmHg was found in 33% of patients with chronic renal failure. the patients with pulmonary hypertension had significantly lower albumin and arteriovenous fistula and long duration of dialysis

Conclusion: This study demonstrated that 42% of patients with chronic renal failure receiving regular hemodialysis have pulmonary hypertension and 24% of patients with chronic renal failure in predialysis have pulmonary hypertension.

The presence of pulmonary hypertension was related to the low level of albumin, presence of arteriovenous fistula and long duration of hemodialysis.

Keywords: Pulmonary hypertension, hemodialysis, predialysis, echocardiography.

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Introduction

Pulmonary hypertension is characterized by elevated pulmonary arterial pressure and secondary right ventricular failure. It is life threatening condition with a poor prognosis if untreated. pulmonary hypertension is defined as a mean pulmonary pressure greater than 25 mmHg at rest or 30 mmHg with exercise as measured by right heart catheterization[1].

Pulmonary hypertension (PH) comprises a group of clinical and pathophysiological entities with similar features but a variety of underlying causes. Many etiologies causing PH have been reported, and one of the background disease seen with patients with PH in chronic renal failure (CRF). however, the pathogenesis of pulmonary hypertension(PH) in this group of patients is not explained satisfactorily[2].

Chronic hemodialysis patients are exposed to continuous pulmonary insults of multifactorial origin. there are several explanations for the development of pulmonary arterial pressure (PAP) in CRF. High cardiac output (CO) resulting from the arteriovenous fistula (AVF) may increase pulmonary artery pressure (PAP) [3].

Metabolic and hormonal derangements caused by chronic renal failure may lead to pulmonary arterial vasoconstriction. Moreover, pulmonary calcification in chronic dialysis patients has been associated with pulmonary dysfunction. Besides, fluid overload and anemia may cause PH. Since 1966, arteriovenous fistula (AVF), developed by Brescia and Cimino, has provided the best vascular

Dept. medicine, Dialysis unit, college of Medicine Al-Nahrain University, Al-Kadhmiya Teaching Hospital.
Adress Correspondence to: Dr. Jawad Kadhem Manuti.
E-mail:drjawadkadhem@yahoo.com
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access allowing long-term hemodialysis increased PAP was found in chronic renal failure patients with surgically created arteriovenous fistula [4]

Patients with PH may develop right ventricular failure with features of systemic venous congestion, pleural effusion, and ascites. it can also result in reduced systemic arterial pressure and intradialytic hypotension[5].

Patients And Methods

The study was performed in AL-Nahrain College of Medicine in AL-Kadhmiya Teaching Hospital during the period of August 2008 to February 2009. One hundred patients (60 male and 40 female) involved in this study of different age group ranging from (10 to 70) years (mean of age 37.3 year) complaining chronic renal failure. 50 patients on regular hemodialysis and 50 in predialysis (conservative treatment).

Exclusion criteria for this patient includes chronic obstructive lung disease, interstitial lung disease, chest wall disease , primary pulmonary hypertension , previous pulmonary embolism, collagen vascular disease, left to right shunt and moderate or sever mitral or aortic valve disease.

Each patient underwent full clinical evaluation with special emphasis on any clinical condition that predisposes to pulmonary hypertension, chest radiography, and pulmonary function tests (PFTs), standard 12-lead electrocardiography (ECG), and echocardiography before enrollment to the study and repeated after 6 month. Two-dimensional and M-mode, Doppler echocardiography was performed in all patients. In the presence of tricuspid regurgitation, continuous-wave Doppler echocardiography was used to estimate the systolic pulmonary artery pressure(PAP). Systolic right ventricular (or pulmonary artery) pressure was calculated using the modified Bernoulli equation: PAP = 4 x (tricuspid systolic jet)^2 + 10 mmHg (estimated right atrial pressure).pulmonary hypertension(PH) was defined as a systolic PAP > 35 mmHg[6].

Patients general data (age, sex, comorbidity, medication used), data regarding the kidney disease (etiology of renal disease, duration of renal failure, duration of hemodialysis therapy) and data pertaining to the arteriovenous fistula (AVF) (duration and location of arteriovenous fistula (AVF) were recorded directly from the patients. Laboratory investigations included blood urea nitrogen, creatinine, serum calcium, phosphorus, hemoglobin, hematocrit and protein

Patients were followed for at least 6 months. During follow-up, patients with end-stage renal disease (ESRD) who were maintained on chronic hemodialysis therapy were dialyzed for 3 hour 3 times per week.

Statistical analysis was performed using chi-square test. at level of significance $\leq$p.o5 regarded as statistically significant.

Results

The echocardiography findings in patients complaining chronic renal failure are presented in Table 1 and 2.

Pulmonary hypertension (>35mmHg) was found in 12(24%) patients in predialysis (on conservative treatment) and 21(42%) patients under hemodialysis procedure.

Pulmonary artery pressure values are presented as in table 1. Mild pulmonary pressure (35---50mmHg) presented in 22% of the patients. While pulmonary pressure more than 50mmHg was finding in 11% patients.

Data on 33 patients with pulmonary hypertension compared with 67 patients without pulmonary hypertension in table 2. There was no significant difference between both

groups, with regard to the sex, hemoglobin level and ejection fraction (p.>0.05).

Albumin level, presence of arteriovenous fistula and duration of hemodialysis associated with high significant pulmonary hypertension (p. <0.05).

All patients had normal baseline cardiac output as expressed by ejection fraction.

Pericardial effusion was observed in 17(34%) patients in predialysis on (conservative treatment) and 28(56%) patients receiving hemodialysis.

Table 1:Systolic pulmonary artery pressure(PAP) in chronic renal failure

	predialysis	hemodialysis	percentage
Total patients	50	50	
PAP ≤35 (mmHg)	38	29	67%
PAP—35--50 (mmHg)	9	13	22%
PAP-- ≥ 50 (mmHg)	3	8	11%

Table 2:Echocardiography finding in patient with chronic renal failure

Abnormal Echocardiography finding	predialysis No.= 50	Hemodialysis No. = 50
Pulmonary artery pressure>35mmhg	11	19
Ejection fraction mean ±SD(%)	57± 6	58 ± 3
Pericardial effusion	34%	56%
Left ventricular pressure overload	74%	86%
Ischemic heart disease	22%	30%

Table 3:Clinical and laboratory data in patient with normal pulmonary arterial pressure (PAP)group 1 versus patient with high PAP group 2

Variables	Group 1	Group 2	P value
Patients number	67	33	
Male/female	32/25	18/15	
Hemoglobin mean ±SD	10 ± 2g/dL	9 ± 2g/dL	>0.05
Albumin mean ±SD	4.6 ± 1.3 g/dL	3.3±0.4g/dL	<0.05
Ejection fraction ±SD	49% ± 4%	47% ± 11%	>0.05
Duration of dialysis	0----2 year	6 month ---2 year	<0.05
Presence of AVF	7	26	<0.05
Systolic blood pressure mmHg	135—180 mmHg	120—190 mmHg	>0.05
Diastolic blood pressure mmHg	70—110 mmHg	75—130 mmHg	>0.05

Discussion

Although there are probably genetic determinants, environmental exposures and acquired disorders that predispose the patients to pulmonary arterial hypertension, it is clear that none of the factors alone is sufficient to activate the pathways essential for the development of this vascular disease. Pulmonary hypertension (PH) involves vasoconstriction and obliteration of the lumen of small vessels in the lungs by plexiform lesions resulting in increased resistance to flow. [7, 8]

In this study, the prevalence of pulmonary hypertension as defined by Doppler echocardiography assessment of tricuspid valve was almost 21(42%) in hemodialysis patients and 12(24%) in predialysis.

The reported prevalence of pulmonary hypertension disease in other study range from 26% to40 %[9]

We compared the clinical and biochemical variables of the patients with different severity and without pulmonary hypertension in chronic renal failure. The patients with pulmonary hypertension had significantly lower albumin, presence of arteriovenous fistula and more than 6 month duration of dialysis (p. value < 0.05). There was no significant deference between hemodialysis and predialysis groups according to ejection fraction, systolic and diastolic pressure, hemoglobin and hematocrit levels (p. value >0.05).

Arteriovenous fistula(AVF), used for hemodialysis access, causes a large left to-right shunt whose capacity often increases with time, and this shunt causes left ventricular failure by imposing an extra volume load on the left ventricle.

In other study, Yigla et al evaluated the incidence of PH in 58 end stage renal disease (ESRD) patients using Doppler echocardiography. Almost 40% of patients had systolic PAP above 35 mmHg, and their cardiac output was significantly higher compared with hemodialysis patients without PH. Because the study population had no obvious cause for PH, they assumed that some factors, such as the size or the location of arteriovenous fistulae are involved in the mechanism that increases cardiac output and contribute to the pathogenesis of pulmonary hypertension(PH)[4].

Pulmonary hypertension (PHT) has an insidious nature and results in extremely serious morbidity. Early detection of the disease is necessary before the development of significant pathophysiological changes. Despite the possibility of common mediators for all the mechanisms of pulmonary hypertension, there are clear differences observed in the potential reversibility of pathophysiological responses of the three components of pulmonary artery pressure that include volume of pulmonary blood flow, resistance in the pulmonary vascular bed and pulmonary venous pressure[10].

Pulmonary hypertension has an insidious nature and results in serious morbidity and mortality so that early detection of pulmonary hypertension is necessary before the development of significant pathophysiological changes.

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Morphometric study on the Ag-NOR changes in skeletal muscle resident cells with aging

May Fadhil Majid Al-Habib *PhD*, **Huda Rashid Kareem** *MSc* .

Abstract

Background: Aging is the deterioration of mature organism resulting from time dependent irreversible changes. The effects of aging on skeletal muscle cells have not been much-elucidated using Ag-NOR analysis.

Objectives: This study aims to demonstrate the effects of aging on Ag-NOR in morphometric & counting aspects.

Materials and methods: The Extensor digitorum longus muscle of forty Albino male rats with age ranging from 27 days up 18 month were studied. Paraffin blocks were performed & sectioned. Ag –Nor stained sections were de-waxed, rehydrted, developer solution was used. Morphometric Analysis of the Ag-NOR stained nuclei through using a soft wear GLI (Global lab image 2) with a microscope connected to PC Unit, a software used to analyze the picture that seen through the microscope, nuclear area, nuclear perimeter, and roundness were calculated,& counting of the number of Ag-NOR stained nucleoli per stained cells.

Results: In neonate age group, the nuclei have high affinity to the stain. High proportion of nuclei can be recognized, with the higher count of Ag-NORs per cells.

- In adult age groups the affinity to the stain is reduced, the nucleus appears to have smaller count of Ag-NORs per cells.
- In old age group the staining intensity seem to be highly reduced, the nucleus seem to have single, rounded Ag-NOR.

Conclusion: A significance differences is seen in Ag NOR_s in cells of skeletal muscle fibers with aging demonstrated by counting and morphometric method of Ag-NOR analysis.

Key words: Skeletal muscle, Ag NOR. Morphometry.

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Introduction

Aging is defined as the deterioration of mature organism resulting from time dependent irreversible changes in all members of species, such that with the passage of time they became increasingly unable to cope with the stress of the environment, and their increasing the probability of death [(1)]. Recent theory of aging based on telomere uncapping at senescence [(2).]

Two aspects of muscle cell phenotype may be affected by aging:

1. The proliferative capacity.
2. The ability to differentiate [(3)].

Dept. Human Anatomy, Section of Histology and Embryology, college of Medicine Al-Nahrain University.
Adress Correspondence to: Dr. May Fadhil Majid Al Habib
Kadhmiya, Baghdad – Iraq
P.O.Box 14222
E-mail: mayalhabib@yahoo.com
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Nucleolar organizer regions (NORs) are loops of DNA, which contain ribosomal RNA genes. They are transcripted by RNA polymerase I, and are of vital importance for ultimate synthesis of protein. Ag-NORs are acidic proteins associated to the NORs, which are selectively stained by silver colloid technique [(4)].

Ag-NORs are related to the proliferative capacity, reflecting essentially cell cycle speed. The increase in their number would signify a shorter time of cell cycle [(5)].

The number of NOR at any given stage of cell cycle appears to be inversely proportional to the cell cycle time, thus the higher the amount of NOR, the shorter the cycle time [(6)], and the faster the rapidity of cell proliferation as recognized by (Brugal in 1994) [(7)].

However, the effects of aging on skeletal muscles have not been much-elucidated morphometricly. This study

aims to demonstrate the effects of aging on Ag-NOR in morphometric aspects.

Materials and methods

Animal housing and sampling:

A sample of forty Albino rats (Rattus norvegicus) of male rats was selected for this study, their age ranging from (27-day) up to (18-month).

They were divided into three age groups:

A. 1st age group (Neonate: 1st month of life).

B. 2nd age group (Adult animals: 6-12 month old).

C. 3rd age group (Old animals: More than 12-month old).

Sacrificed animal were killed with chloroform .Extensor digitorum longus muscle was selected for the study, it was taken out by cutting it from it's both ends, and divided transversely into two halves and put into a fixative [8].

General histological preparation of paraffin blocks:

Fixation done by using fisher fixative for 12 hours sections then Paraffin blocks were prepared [9] & sectioned at (5-6µM) thickness using Richert –Jung 3030 –mot Biocut microtome.

Ag –Nor (Argyrophilic nucleolar organizer region):

Sections were stained with Ag-Nor according to Ploton *et.al* [10].

Morphometric Analysis of the Ag-NOR stained nuclei:

Through using a soft wear GLI (Global lab image 2) in which a microscope is connected to PC Unit, and a software is used that analyze the picture that is seen through the microscope ,and captured to be stored in the digital memory. Displayed by the monitor, and analyzed by the software options. The nuclear area, nuclear perimeter, and roundness were calculated through encircling the nucleus that are stained with Ag-NOR by the marker through using the mouse of the computer, and set the software options, the results were expressed in an excel work sheet. Morphometric analysis of the Ag-NOR stained area had been studied by Lorand- & Carvalho 1998 [11] and was considered a valuable index for the nuclear activity yet, perimeter and roundness coefficient were used for the first time in this study.

Counting of Ag-NOR stained nucleoli:

Considering the enumeration of each silver stained dot per cell directly at the microscope focusing through the section thickness at very high magnification (100X) according to the recommendation of Crocker *et., al* 1989 [12].

Results

All examined sections stained with silver nitrate Ag-NOR stain in this study show adequate staining, but variation in the staining intensity among the different age groups was noticed. In neonate age group, the nuclei have high affinity to the stain ,they stained darkly brown, and the muscle fibers stained light brown, high proportion of nuclei can be recognized per section (Figure 1) the nucleus itself seem to have multiple Ag-NOR stained nucleoli that are small sized, rounded ,solitary, darkly stained structures, can be seen through focusing at the site of the nucleus at a high magnification (Figure4, Table 2) the morphometric analysis of the mean nuclear area show the largest value in neonate, the mean nuclear perimeter also is very high, and the nuclei are nearly rounded in shape since mean roundness value is high (as the value of

roundness approach 1 the structure is nearer to a circle) (Table 1) .

In adult age groups the affinity to the stain is reduced, the nucleus appear as single dot, small proportion of the nuclei can be recognized per section with reduction in the number of Ag-NORs per cell (Figure 2 , Table 2) the mean nuclear area, mean nuclear perimeters are also reduced, and the nucleus is nearly oval rather than rounded .

In old age group the staining intensity seem to be highly reduced, the nuclei are lightly stained ,also the muscle fibers, and smaller proportion of nuclei that can be recognized per section (Figure 3), the nucleus seem to have single ,solitary, rounded Ag-NOR, that can be seen through careful focusing at the site of the nucleus at a high magnification (Figure 5, Table 2) the nuclear area, mean nuclear perimeters are greatly reduced in old age group, and nuclear roundness value is very small (Table 1).

Discussion

In this study, all sections showed adequate Ag-NOR silver staining, the muscle fibers are stained in a light brown color and muscle fiber cells, satellite cells are stained with dark brown color.

The satellite cells considered as the active cell in skeletal muscles and as Ag-NOR staining depends on the transcriptional activity of the cells [(14)]. More recently it was suggested that the amounts of silver stained proteins are related to cell proliferation activity [(15)], so the more rapidly growing the tissue or (cells) , the more Ag-NOR proteins are present [(16)]. While the myonuclei are not stained with Ag-NOR since they are mitoticly inactive [(17).]

Ag-NOR technique has been used to asses the degree of differentiation, since clusters of Ag-NORs are only observed in proliferating cells, and Ag-NOR number decreases with cell maturation [(11)].

The argyrophilia of the nucleolar Ag-NOR is a good cytochemical marker of rRNA and of the level of its transcription, consequently the Ag-NOR stainability give further information on the actual or potentially active substructure of the nucleolus, and allow study of their number [(10)]. Their number, size and distribution within the nucleoli at the level of light might be expected to reflect cellular activity, and proliferation [(18)].

In neonate age group, large proportion of nuclei that stained with Ag-NOR can be seen in the section (Figure 1), and these nuclei posses the larger nuclear area, since the Ag-nor stained nuclear area has a strong linear correlation with the speed of cell proliferation [(19)].

In neonatal age group, it was observed that nuclei had a larger perimeter, and they are nearly rounded in shape. The nucleus posses several small, solitary, rounded to oval nucleoli (Figure 4, Table 2), this can be due to a high proliferation rate in neonate age group ,where there is a higher dispersion of the silver precipitations due to an impairment of their aggregation into nucleoli, giving rise to many small nucleoli as mentioned by (Crocker& Hotstadter) [(12,19)].

In adult age group there is a marked reduction in the amount of nuclei that stained with Ag-NOR in the section(Figure 2), accompanied by a reduction in the Ag-NOR stained nucleoli per cell (Table 2), and in nuclear area, perimeter. Roundness of nuclei is nearly rounded in shape (Table 1).

In old age group there is only single Ag-NOR stained nucleus in the section (Figure3, Table 2). A reduction in the Ag-NOR numbers was noticed with terminal cellular differentiation as

in a leukemia cell line, and with aging of stimulated lymphocytes [20 & 21]. This accompanied with lower value of nuclear area and perimeter, the nucleus of old age group posses only a single nucleolus (Figure 5), as Ag-NOR decreased with cellular maturation [16 & 22].

In old age group the marked reduction in the number of Ag-NOR staining nuclei in the section, due to a reduction in the number of satellite cells and the Ag-NOR stained structure might represent the reserve of satellite cells, where even in the absence of transcription or proliferation there is a basal level for Ag-NOR proteins [23].

Table 1:The values of morphometric analysis of mean area, perimeter, and roundness of Ag-NOR stained nuclei in the four age groups.

Parameters	Neonate	Adult	Old
Mean Area (micron)^2	**8.890629**	**2.90035**	**1.82137**
Mean Perimeter (micron)	**13.71641**	**8.306701**	**7.462056**
***Mean Roundness**	**0.662692**	**0.411671**	**0.331703**

*** Roundness:** a parameter of comparison of nuclear shape, as its value approaching (1), the nucleus is nearly circular, otherwise, it's oval, spindle shape, or other shapes.

****** All measures were done on oil immersion but pictures here on different magnifications for presentation purposes.

Table 2:The total number of Ag-NOR stained nucleoli per cell in the different sections for each age group.

Sections	Neonate	Adult	Old
Section 1	12	5	1
Section 2	12	5	1
Section 3	13	5	1
Section 4	11	6	1
Section 5	10	7	1

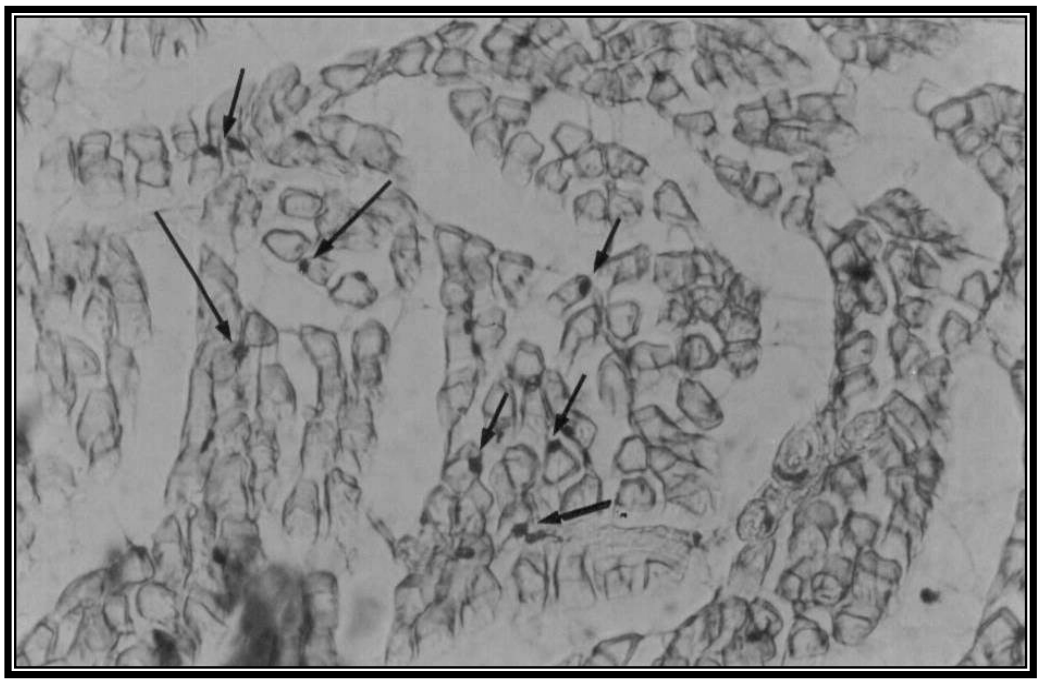

Figure 1: Cross section in skeletal muscle of neonate age group, with high number of nuclei that stained with Ag-NOR silver nitrate stain, the (arrows) demarcate the site of nuclei that have high affinity to the stain. Ag-NOR silver nitrate stain(X800).

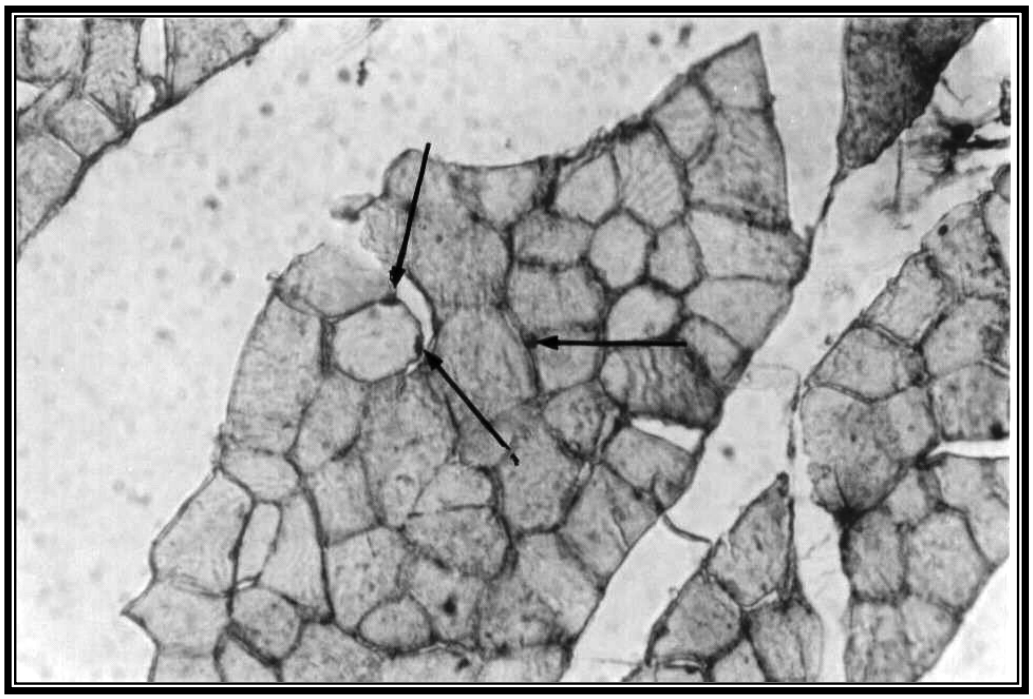

Figure 2: Cross section in skeletal muscle of adult age group, with reduced number of nuclei that stained with Ag-NOR silver nitrate stain (arrows), Ag-NOR silver nitrate stain (X800).

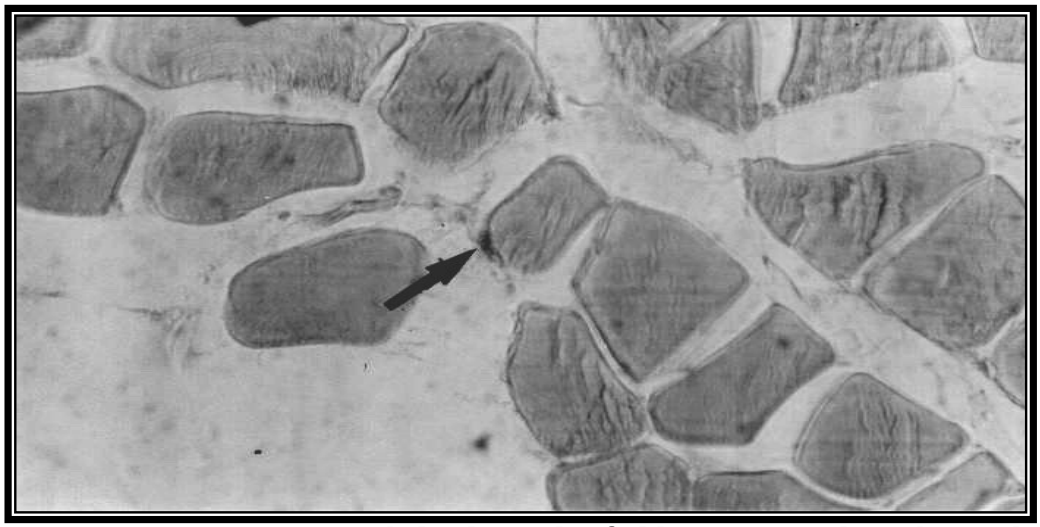

Figure 3: Cross section in skeletal muscle of old age group, show the single nucleus that stained with Ag-NOR silver nitrate stain (arrow), Ag-NOR silver nitrate stain, (X800).

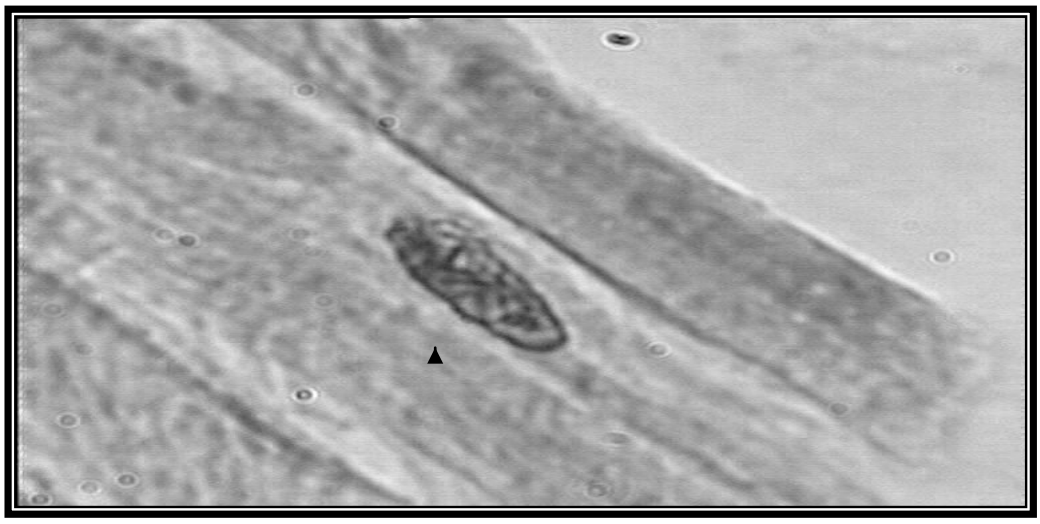

Figure 4: Neonate nucleus with multiple Ag-NORs, Ag-NOR silver nitrate stain, (X2000).

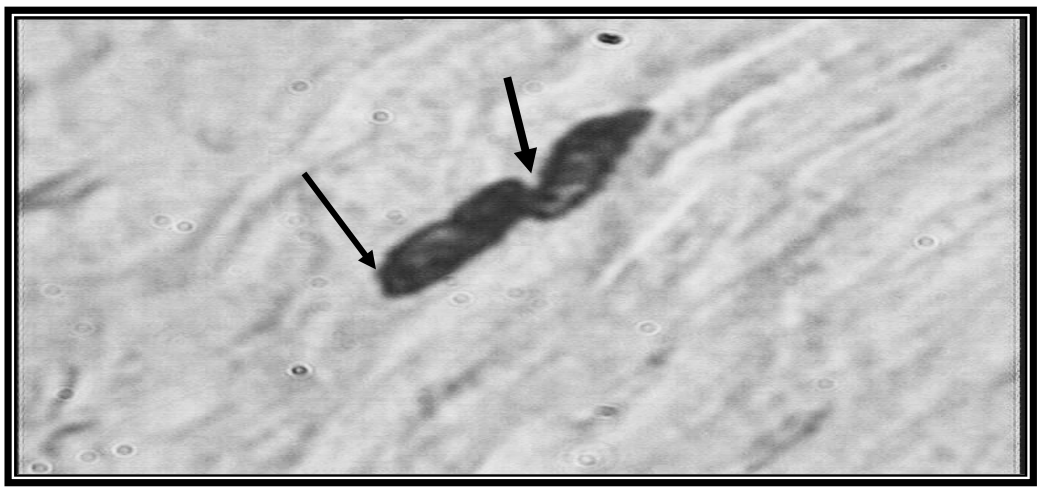

Figure 5: Old age group, show single solitary Ag-NOR per a darkly stained nucleus, Ag-NOR silver nitrate stains (X 2000).

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Trace Elements Homeostasis in Preeclampsia

Faisal Gh. Al-Rubaye *MBChB; MSc ; PhD.*

Abstract

Background: Preeclampsia is a form of high blood pressure with proteinuria manifested during pregnancy, it is a common major complication causing significant morbidity and mortality; however, its etiology is unknown. Moreover, data on mineral and trace elements homeostasis and on cation pattern during pregnancy are conflicting. Also, the status of ionized calcium and magnesium during pregnancy and its complication, preeclampsia, has not been described adequately.

Objective: to demonstrate the pattern of minerals and trace elements during
preeclampsia with respect to normal pregnancy.

Subject and methods: the present study is a cross-sectional case-control study included measurement of minerals (calcium and magnesium) in 60 patients with preeclampsia. They were classified into two groups according to the gestational age:

- Preeclamptics in the second trimester G1: (n=30).
- Preeclamptics in the third trimester G2: (n=30,).

The results are compared with 60 apparently healthy pregnant controls. They were classified according to the gestational age into two groups:

- Pregnants in the second trimester G3: (n=30).
- Pregnants in the third trimester G4: (n=30).

Results: show that serum corrected calcium, serum magnesium and serum zinc were significantly reduced accompanied by significant high serum copper in pre-eclamptics when compared with that of normal pregnants.

Conclusion : preeclamptics (in different gestational age groups) altered mineral and trace element status when compared with healthy pregnants matched with their age and gestational age.

All preeclamptics had certain factors that reduce vasodilation, enhance vasospasm and trigger oxidative stress supported by the finding of low level of ionized magnesium (which is essential for maintenance of vascular tone), low ionized calcium level (which is essential for the synthesis of endothelial-derived NO), high copper level (which generates highly reactive oxygen species) and low zinc level (which is essential for antioxidant function).

Key words: preeclampsia, calcium, magnesium, zinc, copper.

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Introduction

Preeclampsia (PE) is defined as the onset of hypertension and the presence of proteinuria during pregnancy, usually occurring after the 20th week of gestation in a previously normotensive woman and resolving completely by the sixth week after delivery of fetus[1,2].

Dept. Chemistry & Biochemistry, College of Medicine, Al-Nahrain University.
Adress Correspondence to: Dr. Faisal Gh. Al-Rubaye.
E- mail: faisal3ghazi@yahoo.com
Mobile: 07702640792
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The pathophysiology of preeclampsia is thought to represent a defective response to the physiologic demands of normal pregnancy[2,3]. Normal pregnancy is associated with profound changes in maternal homeostasis[4]. The endpoint of these changes is to provide the fetus with the necessary environment for growth and the mother with adequate protection against pregnancy complication[4]. During normal pregnancy, maternal plasma total calcium concentrations fall, primarily because of the decrease in serum albumin to which the mineral is

predominantly bound in the circulation and it seems likely that there is a relatively little change in unbound ionized calcium. However, there is a substantial fetal need for calcium [5]. It is now clear that the dynamics of calcium homoeostasis are in fact substantially altered in pregnancy[5]. Pregnancy-induced hypomagnesemia has been reported previously[6]. However; the status of ionized magnesium during pregnancy and its relation to other important cations such as ionized calcium have not been described adequately [6]. It is the "free" or ionized magnesium that exerts biological activity[6].

It was suggested that a deficiency in magnesium contributed to the development of vasoconstriction in preeclampsia[7]. Also, deficiencies in calcium intake have been linked to preeclampsia/eclampsia, and hypocalciuria and deviations in both 1,25-$(OH)_2D_3$ and PTH have been shown in women with preeclampsia[8].

In addition, endothelium-derived relaxing factor (EDRF) is rapidly inactivated by free radicals. Therefore the hypothetical decrease in EDRF release in preeclampsia might be caused by the increase in oxygen free radical. Because of their biological role as catalyst for endogenous antioxidant enzymes, trace elements have been assessed in many researches to find whether they contribute to the etiology of preeclampsia or not.[9].

Subjects & Methods

A-Subjects

The study was a cross-sectional, case-control study conducted on sixty patients with preeclampsia (PE) attending the Obstetric Consultant-Clinic, Antenatal Clinic, and Labor Ward at Al-Kadhimiya Teaching Hospital, for re-evaluation of newly diagnosed PE, or for delivery.

The diagnosis of PE was based on clinical criteria that were hypertension (absolute BP of 140/90 mmHg twice over 4 hrs without prior comparison)[1,2] and proteinuria (21.5 mg of urinary protein per mmol creatinine)[8] .

The exclusion criteria, which were used for cases and controls, were gestational or chronic hypertension, diabetes mellitus, renal disease, multifetal gestation, intrauterine fetal death, and pregnancy less than 20 weeks of gestation.

Depending on the gestational age, the patients were divided into two groups:

1.Preeclamptics in the second trimester (G1):

Included thirty Preeclamptics in their second trimester of pregnancy. Age range was from 18 to 37 years. The gestational age range was from 20 to 28 weeks.

2.Preeclamptics in the third trimester (G2):

Included thirty preeclamptics in their third trimester of pregnancy. Age range was from 18 to 40 year. Gestational age range was from 29 to 40 weeks.

Controls:

Sixty apparently healthy pregnants attending the Antenatal clinic, and Labor Ward at Al-Kadhimiya Teaching Hospital, for re-evaluation of their pregnancy, or for delivery. The control groups were comparable preeclamptic groups regarding the age, gestational age, Depending on the gestational age, the apparently healthy pregnants were divided into two groups:

3**. Control pregnants in the second trimester (G3):**

They were thirty apparently healthy pregnants in the second trimester of pregnancy. Age range was from 15 to 38 years. Gestational age range was from 20 to 28 weeks.

4. Control pregnants during the third trimester (G4):

They were thirty pregnants in the third trimester of pregnancy. Age range was from 18 to 35 year. Gestational age range was from 29 to 40 weeks.

B. Blood & urine samples:

Ten milliliters of random venous blood were withdrawn from each patient and control, in supine position, without application of tourniquet. Samples then were transferred into clean new plane tube, left at room temperature for 15 minutes for clotting, centrifuged, and the separated serum was transferred into Eppendrof tube, which was used for measuring minerals and trace elements (Ca, Mg, Cu, and Zn). The tubes were stored at -20^{o} C until analysis, which was done within one month after collection [10,11].

Random urine specimens were obtained from each subject in the study to quantify urinary calcium, magnesium, zinc, copper that were expressed as a ratio to urinary creatinine. As a preservative, 1-2 mls of 6M HCl was added to each random urine specimen; the samples were stored in appropriate containers at -20ºC until analysis[10,11].

C-Methods

The assay for calcium and magnesium estimation was carried out using *atomic absorption spectrophotometer* (Buck Scientific 210 JVP)[10, 11].

Corrected serum calcium was calculated according to the formula used by Gowenlock-AH [12]:

Instead of obtaining a crude correction for measured calcium, the same data was used to calculate the *ionized calcium(Cai)* according to the formula used by Gowenlock-AH [12]:

The concentration of *magnesium ion* in serum was calculated from measurement of concentrations of total serum protein and total serum magnesium according to the equation used by Willis-MJ and Sunderman-FW[13].

Results

It was found that *the serum corrected and ionized calcium concentrations* were low in the preeclamptic women in third trimester G2 group as compared to healthy controls in the third trimester G4 [P< 0.001 for both parameters] and even when compared to the preeclamptics in the second trimester G1 [P< 0.001 for both parameters] as it can be seen in Table 1. The same significant reduction in *corrected but not ionized calcium* was noticed in the second trimester group G1 when compared to the healthy pregnants in the second trimester group G3 [P< 0.001 for corrected calcium but greater than 0.05 for ionized calcium] as it can be seen in Table 1. There was no significant difference in *corrected and ionized serum calcium* values between healthy pregnants in each group [P > 0.05 for both parameters].

Although the *urinary excretion of calcium (expressed as urinary calcium per creatinine)* was significantly reduced in preeclamptics in both groups G1 [P< 0.01] and G2 [P< 0.05] in comparison with pregnant controls of the same gestational period G3 and G4, the level of *urinary calcium excretion* was not significantly different from that of preeclamptics in the second and third

trimester ,G1 and G2 , [P > 0.05] as well as healthy pregnants in the same gestational periods , G3 and G4 , [P > 0.05].

A significant reduction in both *total and ionized serum magnesium* was noticed through out the course of pregnancy whether among the preeclamptics groups: G1 and G2 [P < 0.001 for both parameters]; or among healthy control pregnant G3 and G4 [P < 0.001 for both parameters]. When preeclamptic groups G1 and G2 were compared with the corresponding healthy control pregnant groups G3 and G4, the reduction in *total and ionized serum magnesium* was also significant [P < 0.001 for both parameters].

A significant elevation in *urinary magnesium excretion expressed as a ratio of urinary magnesium to urinary creatinine* was noticed through out the course of pregnancy whether among the preeclamptic groups: G1 and G2 [P< 0.001]; or among healthy control pregnant ,G3 and G4 [P<0.01 for both parameters]. When preeclamptic groups G1 and G2 were compared with corresponding healthy control pregnant groups G3 and G4, the significant increase in *magnesium excretion* is also found [P < 0.001].

Serum zinc was significantly lower in preeclamptics (G1 &G2) compared with normal pregnants (G3 & G4) [P < 0.05 for both]. Also serum zinc was significantly lower in G2 compared with G1 [P < 0.05], but there was no significant difference between G3 & G4 [P > 0.05].

Urinary excretion of zinc expressed as zinc: creatinine ratio was significantly increased in preeclamptic in the third trimester G2 when compared to corresponding control G4 [P < 0.05], although this increase excretion of zinc was seen when second trimester groups (G1 & G3) were compared with each other; it failed to reach to a statistically significant level. Moreover, the increment in zinc excretion failed to reach statistically significant level when preeclamptic and pregnant groups were compared with each other [P>0.05].

Serum copper level was significantly higher in preeclamptics (G1 &G2) compared with normal pregnants (G3 & G4) [P < 0.05 for both]. Also serum copper level was significantly higher in G2 compared with G1 [P < 0.05 for both], but there is no significant difference between G3 & G4 [P > 0.05].

According to urinary copper: creatinine ratio, there was a significant retention of copper in preeclamptic in the third trimester G2 when compared with respective control G4 and even with preeclamptic in the second trimester [P < 0.05 for both]. This retention is not significant when preeclamptic in the second trimester G1 was compared with corresponding control G3 [P >0.05] and also among pregnant controls when compared with each other [P > 0.05].

Discussion

A number of studies have found that ***serum total calcium*** level is not different in non-pregnant controls and healthy pregnant women, whereas other researchers like Pederson et. al.[14], found decreased total serum calcium values in normal pregnancy. Furthermore, the beneficial role of a calcium supplementation in preeclampsia is still controversial[15,16]. Some investigators reported an increased free erythrocyte and platelets calcium concentration, speculating that transmembrane calcium fluxes re-altered in hypertensive pregnancy, possibly by a specific mechanism probably of

placental origin[7]. The finding of low serum total calcium in preeclamptics reported here is in agreement with the findings of Ingec et.al.[17], Kisters et.al.[7], Hojo & August[18] who concluded that a calcium deficit leads to an increased intracellular ionized calcium concentration during late pregnancy contribute to the pathogenesis of preeclampsia. In contrary, many investigators like [6, 14, 19-22] found that serum calcium did not differ significantly from normal pregnant group.

Regarding the **ionized fraction of calcium** which is crucial for the synthesis of vasoactive substances in the endothelium as prostacyclin and nitric oxide[23]. The finding of significant reduction in this fraction, as seen in Table 1 is consistent with those reported by Seely et.al.[24], who concluded that a low level of active vitamin D (1,25-$(OH)_2$ D) in preeclamptics, may contribute to suboptimal intestinal absorption of calcium during a time of increased calcium demand resulting in lower ionized calcium, increased PTH, and hypocalciuria in preeclampsia[6]. Abnormalities in calcium homeostasis may contribute to the increased vascular sensitivity documented in preeclampsia. In contradiction to the reported difference in ionized calcium between normal and preeclamptic patients, other authors like [14, 19, 21, 22] found no difference in serum ionized calcium.

Urinary calcium in preeclamptic in this study was observed to be lowered as compared to corresponding control pregnant as seen in Table 1.

The etiology of hypocalciuria in preeclampsia is unknown. However, different assumptions have been given[25]. Particularly, it has been proposed that hypocalciuria may result from decreased dietary intake of calcium resulting in a low circulating calcium and hence low urinary calcium[25]; or from decrease intestinal absorption as secondary result of decreased 1,25 dihydroxyvitamin D, which enhances intestinal absorption of calcium[25]; or it may be due to increased calcium intake by the growing fetus and placenta[25]; lastly, it may be due to intrinsic renal tubular dysfunction, presumably due to decreased glomerular filtration and increased tubular reabsorption[25].

A decrease in both **total and ionized magnesium** was observed through out the course of pregnancy in both normal and preeclamptic pregnant women as seen in Table 1.

Several studies like [6, 7, 26-30] reported that hypomagnesaemia was associated with pregnancy. The level of magnesium cation studied was found to be within the same ranges reported for corresponding non-pregnants in other studies like Brooks & Fry[31], Richard et. al.[22]. other reseachers like Sanders et. al.[19] reported an increase in serum magnesium level in sever preeclampsia. Although the reason for the reduction in total and ionized magnesium is not clear, it is not likely to be due solely to hemodiluton and extracellular fluid volume expansion as serum magnesium levels are still observed to decrease when corrected for protein dilution[6]. An increase in the renal clearance during pregnancy may contribute to the reduction in serum magnesium, since the kidney is the main regulator of the body magnesium[6]. This was supported by the finding of significant increase in magnesium excretion in healthy control and preeclamptic pregnancy with advancing gestational age according to magnesium: creatinine ratio, as it can be seen in Table 1. Other factors that may

contribute to hypomagnesaemia in pregnancy include poor dietary intake[6] which is accompanied by consumption of minerals by the growing fetal skeletal system[6]. Hypoproteinaemia is another contributing factor since extracellular magnesium accounts for about 1% of the total body magnesium content. About 55% of magnesium is free, 30% is associated with proteins (primarily albumin), and 15% is complexed with phosphate, citrate, and other anions[10]. The technique used for measuring ionized magnesium can also be considered, ideally, it is the ion-selective electrode, instead a mathematical equation was employed[10,12].

The relation between serum total and ionized magnesium with intracellular magnesium has not been defined clearly. In previous study[32], there was no significant difference in red blood cell magnesium levels in teenagers with pregnancy-induced hypertension, whereas plasma magnesium tended to decrease with increasing gestation in this same group. However, recent evidence suggests that extracellular magnesium may modulate intracellular magnesium in vascular smooth-muscle cells[6].

Signicant changes in serum trace metal concentrations, particularly zinc and copper, have been documented during normal pregnancies[11]. In preeclampsia, decreased maternal and fetal serum zinc levels have been determined. In preeclampsia, the lowered serum zinc concentrations have been suggested to be at least partially due to reduced estrogen and zinc binding protein levels. Serum cortisol level increases during normal pregnancy and it is much higher in preeclampsia, which again reduce maternal zinc levels and subsequent urinary zinc[11]. Several investigators have noted that women with preeclampsia as compared with normotensive pregnant women had lower zinc concentrations[9]. On the other hand, reports on changes in copper levels were conflicting: decreased[33] elevated[34] and unchanged levels[35].The possible causes of these changes are discussed in view of the hormonal, metabolic and enzymatic changes in preeclampsia[9]. The physiological increase in copper concentrations in pregnancy is, in part, associated with estrogen induction of copper carrying protein[9]. In this study serum zinc levels were significantly lower and serum Cu levels were significantly higher in both groups with preeclampsia when compared with normal pregnant groups.

Biochemical changes in preeclampsia appear to involve mineral and trace metal metabolism leading to the appearance of the typical pattern which may cause vasospasm of eclampsia. These changes would include low serum ionized calcium, low serum total and ionized magnesium, low serum zinc with elevated serum copper and imbalance in the urinary excretion of calcium, magnesium and trace elements. Further study of intracellular minerals, oxidant-antioxidant status and the membrane Na, K ATPase and calcium pumps to explore their potential role in the pathogenesis of preeclampsia is required for future work.

Table 1: The mean value of minerals (corrected Ca^{+2}, ionized Ca^{+2}, total Mg^{+2}, ionized Mg^{+2}, ratio of ionized Ca^{+2}: ionized Mg^{+2}) & trace elements in the sera & urine of different preeclamptic and pregnant control groups (presented as mean ± SD).

Variable	G1	G2	G3	G4
Serum corrected calcium (mmol/L)	2.3 ± 0.05	2.2 ± 0.09	2.5 ± 0.1	2.5 ± 0.1
Serum ionized calcium (mmol/L)	1.2 ± 0.08	1.1 ± 0.05	1.2 ± 0.05	1.2 ± 0.05
Urinary calcium : creatinine	0.6 ± 0.27	0.58 ± 0.59	0.94 ± 0.6	0.8 ± 0.19
Serum magnesium (mmol/L)	0.81 ± 0.04	0.65 ± 0.08	1.02 ± 0.04	0.97 ± 0.05
Serum ionized magnesium (mmol/L)	0.45 ± 0.03	0.32 ± 0.06	0.62 ± 0.04	0.43 ± 0.03
Urinary magnesium: creatinine	0.07 ± 0.003	0.09 ± 0.03	0.0149 ± 0.0101	0.04 ± 0.01
Serum Zinc (mmol/L)	0.20±0.019	0.18±0.022	0.24± 0.045	0.22 ±0.042
Serum copper (mmol/L)	0.23± 0.029	0.35± 0.021	0.19± 0.018	0.2 ± 0.028
urinary zinc: creatinine	0.0026 ± 0.004	0.003 ± 0.002	0.0025 ± 0.003	0.0025 ± 0.003
Urinary copper: creatinine	0.006 ± 0.005	0.004 ± 0.005	0.0092 ± 0.007	0.009 ± 0.001

(G1):Preeclamptics in the second trimester.
(G2): Preeclamptics in the third trimester.
(G3):Control pregnants in the second trimester.
(G4):Control pregnants during the third trimester.

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Incidental Intracranial Tumor: A Case Report

Mutaz Abdul Majeed Al-Qazzaz *FICMS*.

Abstract

This is a case report of 30 years lady referred by the investigation authority to the medico-legal institute in Baghdad as a car accident victim for postmortem examination.
A prior autopsy history with her relatives was negative. During autopsy a large intracranial tumor was discovered at the base of the brain. Histopathological examination revealed the diagnosis of meningioma.

Keywords: Intracranial tumor, meningioma, autopsy, brain tumor.

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Introduction

Tumors of the CNS have a unique characterstics that other neoplasms else where in the body don't posses. The distinction between benign and malignant types are less evident with limited regain of neurological functions after surgical resection as well as their fatal capability depends on anatomical site irrespective of histological type [1]. Glioblastoma multiformi is the most common malignant adult brain neoplasm, occurs most frequently in the 5th and 6th decade of life [2].

Meningiomas are the most common benign primary brain tumor discovered incidentally more frequently in elderly persons [3, 4]. They comprise 20% of all intracranial tumors in adults [5].
They arise from the meningiothelial cells of the arachnoid and vary in their size from a pinhead to the size of a man' fist depending on their location, type of growth and growth rate (mostly slowly growing tumors) [6].

Meningiomas are occasionally discovered as incidental findings on CT scan or MRI [7].

Dept. Pathology and Forensic Medicine, college of Medicine Al-Nahrain University.
Adress Correspondence to: Dr. Mutaz A.Al-Qazzaz.
E-mail: alqazzazmutaz58@yahoo.com
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Most meningiomas remain asymptomatic throughout life which explains why 50% of all meningiomas are discovered at autopsy [8]. They are commonly seen in individuals between 3rd and and 6th decade of life with female to male ratio 2:1 [2]. Most common sites of involvement include parasagittal aspect of the brain convexity, dura over the lateral convexity [1] and wing of the sphenoid [9].

Symptoms of the tumors depend on their location, type and rate of growth. They can be highly fatal if they are very large or causing increased intracranial pressure, sever cerebral edema or herniation [2].

Sometimes they can reach an enormous size while producing minimal symptoms especially in the frontal lobe [5].

Objective of this case study

To draw the attention for the presence of some silent and sometimes serious brain pathology and the importance of full investigation even in minimal symptoms.

Case Study

This is a case report of a 30 years old single female brought by the police to the medico-legal institute in Baghdad as a car accident victim. Information regarding the circumstances of her death was gained from the police report as well as from

an interview with her brother who denied any previous medical or surgical history of the deceased.

External examination of the body revealed multiple brush abrasions 1-7 cm in diameter on the anterior right side of the chest and on the external aspects of her upper limbs. A bruise was seen on her upper abdomen of about 10 cm in length.

Dissection of the scalp reveals no abnormal findings.

After removing of the calvarium the brain was seen slightly edematous with congestion. On trying to remove the brain a big mass was noticed at the base of the brain on the left side of the anterior cranial fossa. After removing the brain from the skull the mass was excised. A compression effect by the mass was seen on the basal aspect of the left frontal lobe and to lower extent the temporal lobe.

The Base of the skull was normal.

Evisceration of chest organs was done. Examination of the chest cage showed fractures of the 4^{th}, 5^{th}, 6^{th}, 7^{th} ribs of the right side along the lateral axillary line with bruises at the fractures sites. Right lung showed multiple bruises 5-7 cm on the 3 lobes. A huge abdominal collection of blood intraperitonialy was collected and measured to be almost 3 liters. Multiple tears were seen in the liver which explains the source of blood. Other abdominal and pelvic organs were normal.

Gross examination of the brain mass showed a solid vascular growth weighting 60 gram measuring 5x3x2 cm, hemispherical in shape.

Fixation of the mass was done using 10 % formalin over night.

Histopathological examination using H & E stain revealed transitional type of meningioma. Interrogation with the deceased brother after completion of autopsy examination and facing him with the brain pathology seen during autopsy, he admitted that she was complaining form some symptoms like headache, blurring of vision, mental loss and occasional seizure attack for which she was on antiepileptic medications.

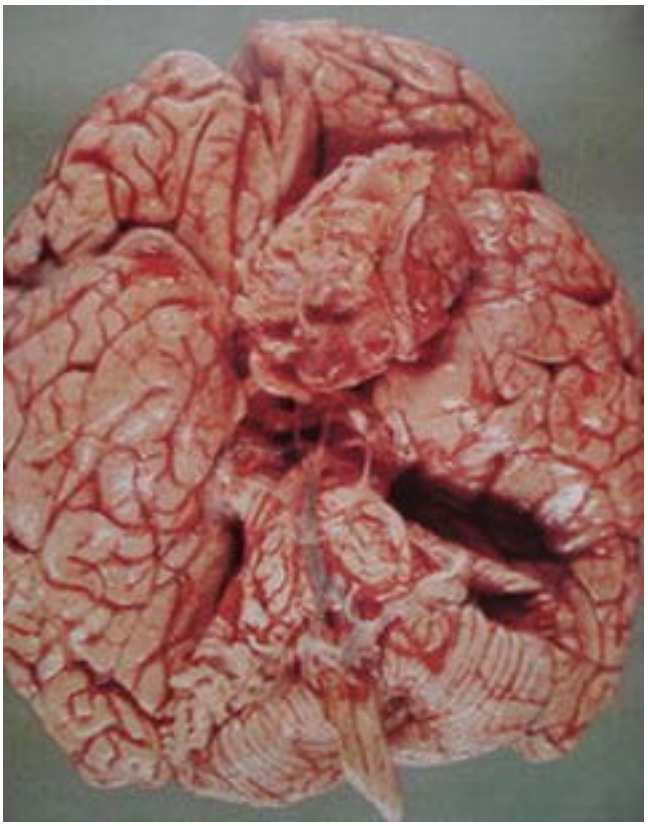

Figure 1: Brain, basal view showing large meningioma at the base of the brain.

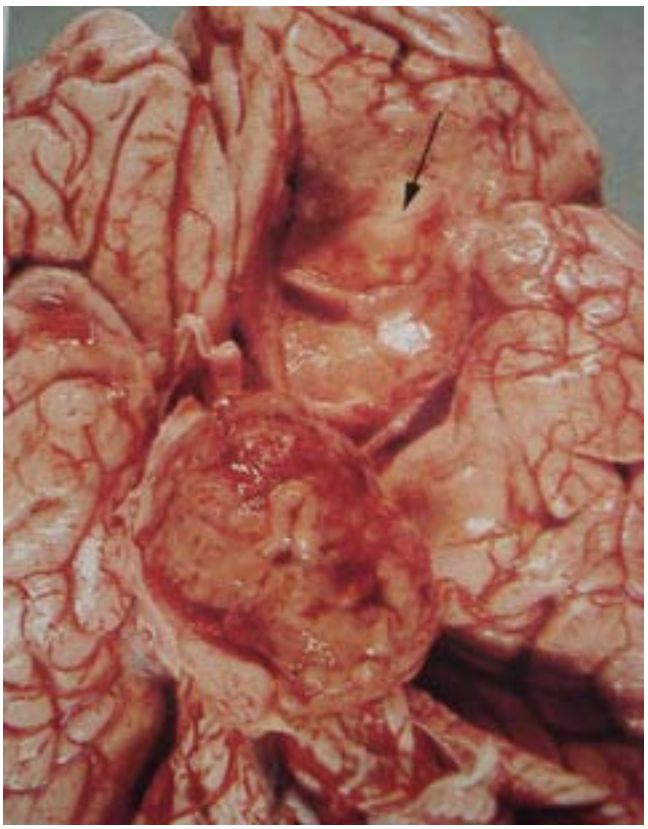

Figure 2: Brain, basal view, showing compression of the underlying structures by the tumor.

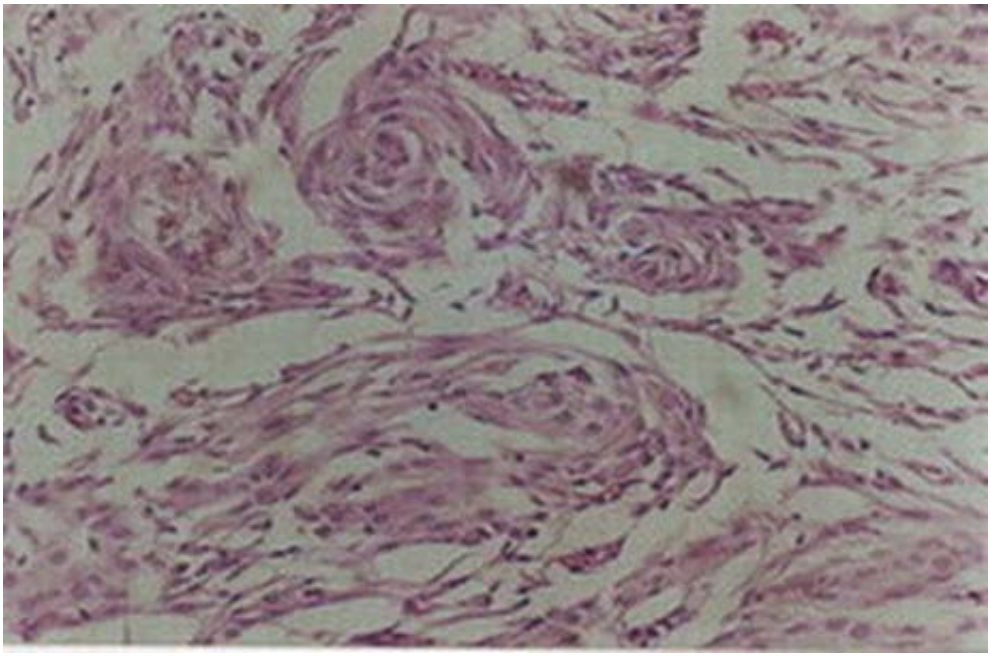

Figure 3: Section from the tumor tissue stained by H & E stain shows whorls of transitional meningothelium (X 40).

Discussion

Brain tumors can affect people of all ages. Meningiomas are benign slowly growing encapsulated highly vascular intracranial tumors [10, 11]. In the current study the age and sex of the deceased coincide with their comparable parameters in previous studies which stated that meningiomas affect mostly between 3d and 6th decade of life more frequently in the age 21-30 years with predilection to female [2,5,6].

Incidental detection of meningioma is more frequently in elderly people than in young people because of the process of brain atrophy as well as calcification of the tumor which lessens the rapidity of growth [3].

In general meningiomas remain asymptomatic throughout life which explains why 50% of all meningiomas are discovered at autopsy [8, 12].

In this study the victim was complaining of some neurological symptoms during her life which forced her to consult a doctor who gave her medications probably without thorough investigations like CT scan or MRI. Those symptoms arised as a result of pressure effect of the tumor on the basal aspect of the frontal lobe as well as the temporal lobe until she died by a road traffic accident.

As far as the site and the size of the tumor are concerned, neither of them were so serious as to cause her death in my opinion as it was reported previously that the size can reach to an enormous one while clinically producing minimal symptoms specially in the frontal lobe [5] as it was in this study, nor the site was as serious as meningioma of the cerebellopontine angle.

Acknowledgment

I would like to express my sincere thanks to Prof.Dr.Yarub I.abdulkader at the dept. of pathology and forensic medicine, college of medicine, University of Al-Nahrain for his assistance in histopathological diagnosis of the tumor.

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المجلة العراقية للعلوم الطبية

قائمة المحتويات

المقالات

المجلد السابع، العدد الثاني، 1430 هـ، 2009م

المجلة العراقية للعلوم الطبية

تعنون المراسلات إلى المجلة العراقية للعلوم الطبية، صندوق بريد 14222 بغداد، العراق. تلفون و فاكس (964-1-5224368).

رقم الإيداع في دار الكتب و الوثائق ببغداد 709 لسنة 2000

الهيئة الأستشارية

أ.د.محمد حسن العلوان(الجامعة المستنصرية)

أ.د.محمود حياوي حماش(جامعة مؤته)

أ.م.د.مزاحم قاسم الخياط(جامعة الموصل)

أ.د. مهـــا محمد جاسهـــم البياتي (جامعة النهرين)

أ.د.نزار الحسني (الهيئة العراقية للأختصاصات الطبية)

أ.د. هاشهـم مهـــدي الكاظمـــي (جامعة النهرين)

أ.د. يعرب إدريس عبد القادر (جامعة النهرين)

توازن المعادن لدى الحوامل المصابات بارتفاع ضغط الدم(قبل الشنج)

فيصل غازي الربيعي[1] , مها البياتي[2],طارق حفظي الخياط [3]

الخلاصة:

خلفية الدراسة: ضغط الدم العالي لدى الحوامل (بريِ إكلامبسيا أو قبل الشنج) هو نوع من أرتفاع ضغط الدم يظهر أثناء الحمل, وهو من المضاعفات الشائعة المؤدية الى نسبة وفيات ومضاضة عاليتين؛ ومع ذلك سبب هذا الارتفاع غيرمعلوم. المعطيات حول أتزان معادن الدم وخاصةالمعادن الموجبة أثناء الحمل تعتبر متناقضة؛ أضافة لذلك, فأن حالة آيونات الكالسيوم والمغنيسيوم الحرة خلال الحمل ومضاعفاته (أرتفاع ضغط الدم المصاحب للحمل " بريِ إكلامبسيا أو قبل الشنج") لم توصف بشكل كامل.

هدف الدراسة: الهدف من هذه الدراسة هو لبيان نمط المعادن (الكالسيوم و المغنيسيوم) في حالة ارتفاع ضغط الدم المصاحب للحمل (بريِ إكلامبسيا أو قبل الشنج) وعلاقته مع الحمل الطبيعي.

المرضى وطرق البحث: هذه الدراسة هي دراسة مقطعية تشمل قياس المعادن (الكالسيوم, المغنيسيوم) لدى 60 حاملا" مصابة بارتفاع ضغط الدم المصاحب للحمل (مجموعة الأختبار) وتم تصنيفهم الى مجموعتين حسب عمر الحمل:

- حوامل مصابات بارتفاع ضغط الدم المصاحب للحمل (قبل الشنج) خلال الفصل الثاني من الحمل (العدد30 مريضة).
- حوامل مصابات بارتفاع ضغط الدم المصاحب للحمل (قبل الشنج) خلال الفصل الثالث من الحمل (العدد30 مريضة).

تم مقارنة النتائج مع نتائج 60 حاملة" سليمة" ظاهريا"(مجموعة السيطرة)، وتم تقسيم مجموعة السيطرة اعتمادا" على عمر الحمل الى مجموعتين:

- حوامل اصحاء ظاهريا" خلال الفصل الثاني من الحمل (العدد30 مريضة).
- حوامل اصحاء ظاهريا" خلال الفصل الثالث من الحمل (العدد30 مريضة).

النتائج: أظهرت النتائج التغيير في شوارد المصل لدى الحوامل ذوات ضغط الدم العالي المصاحب للحمل (قبل الشنج) يشمل انخفاض معنوي لمستوى المغنيسيوم والكالسيوم المصحح للألبومين في مصل الحوامل المرضى لدى مقارنتهم بمجاميع السيطرة المناظرة، مع انخفاض في تركيز الكالسيوم الحر و المغنيسيوم الحر، مع ارتفاع معنوي في نسبة الكالسيوم الحرالى المغنيسيوم الحر.

الأستنتاج: مما تقدم يمكن الأستنتاج أن الحوامل ذوات الضغط العالي المصاحب للحمل (قبل الشنج) يعانون من اختلال حالة توازن معادن الدم عند مقارنتهم مع الحوامل الأصحاء المناظرين للمرضى في العمر وعمر الحمل.

مفتاح الكلمات: قبل الشنج، الكالسيوم، المغنيسيوم.

[1] فرع الكيمياء و الكيمياء الحياتية [كلية الطب _ جامعة النهرين]
[2]فرع الامراض النسائية والتوليد [كلية الطب _ جامعة النهرين]
[3]فرع الكيمياء الحياتية, [كلية الطب _ جامعة بابل]

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استخلاص وتنقية بروتينات الغشاء الخارجي من عزله محليه لبكتريا الـ Klebsiella pneumoniae

عامر هاني رازق[1] , عصام فاضل ألجميلي[2] , نضال عبد المهيمن[1]

الخلاصة:

خلفيه الدراسة: توجد بروتينات الغشاء الخرجي (البورينات) بكميات كبيره في الغشاء الخارجي للبكتريا السالبه لصبغة الكرام حيث تكون قنوات مملوءة بالماء تسمح بتنافذ الجزيئات المحبه للماء خلال الغشاء الخارجي للخلايا البكتيريه. تقسم البورينات بصوره عامه الى بورينات لا نوعيه (مثل OmpF,OmpC) التي تسمح بتنافذ الجزيئات القطبيه الصغيره (600 Da) وبورينات نوعيه (مثل LamB) والتي تسهل تنافذ مواد اساسيه لعمل الانزيمات.

هدف الدراسة: تنقية وتوصيف بروتينات الغشاء الخارجي (البورينات) من عزله محليه من بكتريا ال Klebsiella pneumoniae.

طريقة العمل: استعملت احدى عزلات بكتريا ال Klebsiella pneumoniae المحليه كمصدر لعزل وتنقية بروتينات الغشاء الخارجي. تم تنقية وتوصيف بروتينات الغشاء الخارجي وتم كذلك التحري عن وجود متعدد السكريد الشحمي في المستحضر النهائي باستخدام طريقة الكشف عن حامض الثايوباربجوريك.

النتائج: احتوى المستحضر النهائي على بورينات وبنسبة 3.2 mg/ml. واظهرت نتائج عملية الترحيل الكهربائي ظهور البورينات بشكل حزمتين ذواتي وزنين جزيئيين هما 35 kDa, 36 على التوالي.

الاستنتاجات: تنتج العزله المحليه لبكتريا ال Klebsiella pneumoniae قيد الدراسه بورينات باوزان جزيئيه مشابهه لما تنتجه البكتريا الاخرى السالبه لصبغة الكرام وتنتج نفس العزله نوعين من البورينات تحت الضروف المختبريه القياسيه.

مفتاح الكلمات: البورينات, حامض الثايوباربجوريك, كروموتوكرافيا الهلام, كيتودياوكستنيت.

[1] فرع الأحياء المجهرية [كلية طب النهرين _ جامعة النهرين]

[2] معهد الهندسة الوراثيه والتقنيه الاحيائيه _ جامعة بغداد

المجلة العراقية للعلوم الطبية 2009 م المجلد 7 العدد 2 ص12-17

تقييم دور تغير شكل كريات الدم الحمراء في معدل تجمع وترسب كريات الدم الحمراء باستخدام أشعة الليزر

رويدة عبد الأمير الخزرجي

الخلاصة:

خلفية الدراسة: تجمع كريات الدم الحمراء ظاهرة فسيولوجية مهمة في الدورة الدموية هذه الظاهرة تمثل الخصائص الاساسية للدم الطبيعي والتي تلعب دور مهم في الجهاز الوعائي القلبي, وخاصة في الاوعية الدموية الشعرية .

هدف الدراسة: لتقييم دور قابلية تغير شكل كريات الدم الحمراء على تجمع وترسب هذه الكريات.

طريقة العمل: أجريت الدراسة الحالية على (32) شخص سليم. طريقة اشعة الليزر النافذة استخدمت لهذه الدراسة. ويتم حساب شدة أشعة الليزر النافذة بشكل مستمر خلال عملية التجمع والترسب. تم حساب قيم مختلفة لقابلية تغير شكل الكريات الحمراء وتقييم تأثيرها على كل مرحلة من مراحل التجمع, تكون اللفة و تكون التجمع بمتجه او ثلاث متجهات.

النتائج: تم التعبير عن قيم التغير بشكل الكريات الحمراء بعامل الصلابة, عامل الصلابة العالي لكريات الدم الحمراء يقلل معدل التجمع و معدل التجمع بثلاث متجهات بشكل ملحوظ اكثر من عامل الصلابة المتوسط .

الاستنتاج: قيم مختلفة لقابلية تغير شكل كريات الدم الحمراء, من القليلة الى المتوسطة و من المتوسطة الى العالية, بينت تأثيرات مختلفة على مراحل التجمع و الترسب لهذه الكريات.

مفتاح الكلمات: تجمع كريات الدم الحمراء، معدل الترسب, قابلية التغير بالشكل، أشعة الليزر

فرع الفسلجة [كلية الطب _ جامعة النهرين]

المجلة العراقية للعلوم الطبية 2009 م المجلد 7 العدد 2 ص18-25

السستينيوريا لدى مجموعة من الاطفال في العراق

شذى حسين علي

الخلاصة:

خلفية الدراسة : السستينيوريا مرض وراثي بصفة متنحية ناتج عن خلل في امتصاص الاحماض الامينية الثنائية. زيادة افراز السستين في الادرار لانه الاقل ذوبانا يؤدي الى تكوين الحصى.

هدف الدراسة: نقدم تجربتنا في معالجة السستينيوريا لدى مجموعة من الاطفال في العراق.

طريقة العمل: من 1999 لغاية 2006 , تم تقييم ومعالجة ومتابعة كل الاطفال المصابين بالسستينيوريا في مستشفى الكاظمية التعليمي.

النتائج: كان عدد المرضى المعالجين 23 (16 ذكر, 7 اناث). تراوح معدل العمر من 10 شهور الى 18 سنة .

ظهرازدياد أفراز اليورات في 30.5% , أزدياد أفراز الكالسيوم في 13% , وأزدياد افراز الاوكسالات في 4.3% من المرضى.

المدى الزمني لمتابعة المرضى 1 – 88 شهر.

تسعة مرضى عولجوا بزيادة السوائل والقلويات فقط . تم استعمال عقار البنسلامين في 12 مريض. الاعراض الجانبية للبنسلامين ظهرت لدى 4 مرضى (22.2%) . أعطي عقار الكابتوبريل لاربعة مرضى . تم استعمال تفتيت الحصى في 8 مرضى و 18 مريضا أجريت لهم العمليات الجراحية .

معدل الخلو من الحصى كان 55.6% مع السوائل والقلويات , 58.3% مع البنسلامين , 0% مع الكابتوبريل و 50% مع التفتيت .

وجدت الحاجة للعلاج الائتلافي في 45% من المرضى . كان معدل رجوع الحصى 70% .

الاستنتاج: السوائل والقلويات اكثر نجاحا في حالة استعمالها في الحالات البسيطة.

نتائج العلاج الناجح كانت متساوية حين استعمال البنسلامين والتفتيت .

مفتاح الكلمات: السستينيوريا, الاطفال, الحصى, حصى المجاري البولية

فرع طب الاطفال [كلية طب _ جامعة النهرين]

المجلة العراقية للعلوم الطبية 2009 م المجلد 7 العدد 2 ص26-34

التعبير عن المعلمCD30 في مصول وعلى الخلايا التائيه في نسيج المشيمه الماخوذ من مريضات الاجهاض التلقائي المتكرر

نضال عبد المهيمن[1], امال حسين[2]

الخلاصة:

خلفية الدراسة: من الممكن تحييد الاستجاب المناعيه خلال فترة الحمل التفاعلات تتضمن النوع الثانى من الخلايا التائيه المساعده خاصة فى الموقع الامى الجنينى البينى. و قد عرف بان المعلم الخلوىCD30 يعبر عنه من قبل هذا النوع من الخلايا.

هدف الدراسة: تهدف الدراسه الى التحرى عن مستوى المعلم CD30فى مصول والخلايا التائيه الموجوده فى النسيج المغذىللجنين.

المواد و طرائق العمل: تضمنت الدراسه الحاليه احدى وستون إمرأة، تراوحت متوسط اعمارهن بين (23.9– 28.5)، تم تقسيمهن الى ثلاثة مجاميع: مجموعة(أ) إجهاض تلقائي متكرّر (RSA) وعددهن 35 إمرأة و متوسط اعمارهن بينَ (28.5 ± 0.68). مجموعة (ب) - إجهاض تلقائي غير متكرّر (non-RSA) وعددهن 16 إمرأة و متوسط اعمارهن بينَ (26.4 ± 0.85). مجموعة(ج) - سيطرة (حمل ناجح): وعددهن 10 نساء ومتوسط اعمارهن بينَ (23.9 ± 0.88).. تم جمع نماذج دم من كل المرضى وكذلك مجموعه السيطره. درس المستوى المصلى للمعلم الذائبCD30باستخدام تقنية الاليزا كما جمعت نماذج النسيج المغذي للجنين (التروفوبلاست) من كل المرضى وكذلك مجموعه السيطره لدراسة المعلم باستخدام التحليل المناعي النسيجي الكيميائي (IHC) .

النتائج: اظهرت نتائج حساب مستويات CD30 في النسيج و حساب تركيز sCD30 في المصل وجود زياده معنويه ($p<0.01$) بالنسبة لمتوسط النسبة المؤية ل CD30 عند مقارنة كل من المجموعة (أ) والمجموعة (ب) مع المجموعة (ج) باستعمال طريقتي الفحص .

الاستنتاجات: يعتقد بأنه CD30 يرتبط بخلايا Th2 وبذلك يظهر دوره في الحمل الطبيعي الناجح سواء في نسيج التروفوبلاست او تركيزه في المصل.

مفتاح الكلمات: إجهاض تلقائي متكرّر, تقنية الاليزا , التحليل المناعي النسيجي الكيميائي

[1] فرع الأحياء المجهرية [كلية طب _ جامعة النهرين]

[2] فرع الأحياء المجهرية [كلية طب _ الجامعة المستنصرية]

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طبيعة الممانعة الدوائية لحيوانات التدرن عند المرضى المعالجين سابقاً في العراق

مصطفى نعمه عبد علي , هاشم مهدي هاشم الكاظمي

الخلاصة:

خلفية الدراسة: تعدّ الممانعة الدوائية لعصيات التدرن من الظواهر التي تناولتها البحوث الطبية من زمن بعيد وهي عبارة عن تفاقم الظاهرة الطبيعية لممانعة العصيات بفعل الانسان. وقد سجلت منظمة الصحة العالمية في سنة 1995 خمسين مليون انسان مصاب بهذه العصيات الممانعة. تقسم الممانعة الدوائية بالنسبة لمرض التدرن الى ثلاثة اقسام: الممانعة عند الحالات المرضية الجديدة والممانعة عند المرضى الذين تناولوا العلاج لمدة شهر (في اي فترة سابقة) والممانعة المشتركة. وقد ثبت من خلال الدراسات ان الطفرات الوراثية الحاصلة للعصيات مسؤولة عن هذه الممانعة وان تناول الدواء المضاد للتدرن بصورة غير منظمة من المؤشرات المهمة لحدوثها .

هدف الدراسة: 1- معرفة مقدار الاصابة بالممانعة الدوائية لعصيات التدرن في العراق 2- مقارنة طبيعة الممانعة الدوائية في العراق مع دول اخرى.

طريقة العمل: تم شمول 411 مريضا مصابين بالتدرن في هذه الدراسة كانوا قد راجعوا معهد التدرن في بغداد بشرط ان يكونوا معالجين سابقا لمدة شهر كحد ادنى. وقد تمت الدراسة في الفترة من شهر شباط 2005 الى شهر اب 2006 وتم ارسال عينات القشع الى المختبر لاجراء الزرع الجرثومي الخاص بالتدرن وبعد ان يثبت وجود العصيات يتم فحص الحساسية الدوائية ثم تسجل النتائج.

النتائج: عدد الذكور 311 والاناث 100بمتوسط عمر 34 سنة ونسبة الذكور الى الاناث 1:3 ،كانت189(48.2%) من الزروعات متحسسة لـ: الايزونيازايد،الريفامبسين،الستربتوماسين والايثامبيوتول و213 (51,8%) ممانعة لواحد من الادوية الاربعة على الاقل. وقد شكلت الممانعة ضد عقار الريفامبسين24,4% متغلبة على باقي انواع الممانعة المنفردة. والممانعة المتعددة وجدت عند 52 حالة (24,4%)،اما الممانعة المشتركة للريفامبسين مع الستربتومايسين ومن جهة اخرى الايزونيازايد مع الستربتوماسين فقد وجد انها اكثر انواع الممانعة المشتركة فقد شكلت 9,4% و 4,7% على التوالي. اما الممانعة الدوائية ضد الريفامبسين باي شكل من اشكال الممانعة فقد تغلبت على كل الانواع الاخر للممانعة فقد وجدت عند 146 حالة (68.5%).

الاستنتاج: ان الممانعة الدوائية لعصيات التدرن في العراق موجودة بنسبة كبيرة متغلبة على العديد من البلدان المجاورة ،وان الممانعة الدوائية ضد الريفامبسين (العقار ذو الفعالية العالية جدا ضد التدرن) قد فاقت بقية انواع الممانعة ضد الادوية المضادة للتدرن في حالات المرضى المعالجين مسبقا.

مفتاح الكلمات: التدرن،الممانعة الدوائية لمضادات التدرن،المانعة الدوائية المتعددة.

فرع الباطنية [كلية الطب_جامعة النهرين/ مستشفى الكاظمية التعليمي]

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دراسة التنميط المناعي للخلايا اللمفاوية في الدم المحيطي للأشخاص المتعرضين للمجال الكهرومغناطيسي

رافد عبد الواحد

الخلاصة:

خلفية الدراسة: اثبتت الكثير من الدراسات والبحوث الميدانية الاثار السلبية للمجالات الكهرومغناطيسية والمتولدة من ابراج الضغط العالي لخطوط نقل الطاقة الكهربائية,وخصوصا التاثير على الجهاز المناعي للانسان حيث ان المجال الكهرومغناطيسي يؤدي الى حدوث خللا واضحافي مكونات الجهاز المناعي ومنها التاثير في عمليات التكاثر والتمايزللخلايا المناعيةاضافة الى التداخل مع الاتصالات والاشارات الخلوية للخلايا المناعيةمما يؤثر سلبا في وظيفة تلك الخلايا المناعية.

هدف الدراسة: التحري عن تأثير المجالات الكهرومغناطيسية والمتولدة من ابراج الضغط العالي لخطوط نقل الطاقة الكهربائية على الخلايا اللمفاوية للدم المحيطي للاشخاص المتعرضين لتلك المجالات .

المواد وطرق العمل: أجريت هذه الدراسة المناعية على 60 عينة دم تم اختيارهم بشكل عشوائي 45 شخصا منهم متعرضين للمجال الكهرومغناطيسي الناتج من ابراج الضغط العالي لخطوط نقل الطاقة الكهربائية في مناطق سكناهم حيث تم اختيار النماذج من ثلاثة مناطق مختلفة من بغداد تضمنت البلديات وحي العدل والدورة كذلك تم اختيار 15 شخصا غير متعرضين كمجموعة سيطرة .

تضمنت الدراسة تحديد المعلمات المناعية لخلايا الدم اللمفاوية المحيطية باستعمال اختبار الوميض المناعي المباشر

(imunofluorescence) حيث تم في هذه الدراسة استخدام الأجسام المضادة للخلايا المفاوية التائية التي تحمل المعلم المناعي CD3,CD4,CD8,CD21,CD56 كذلك تم الكشف عن النسبة المئوية لـ CD4/CD8 .

النتائج: اظهرت النتائج وجود انخفاض معنوي واضح في الخلايا التي تحمل المعلم المناعي (CD3, CD4, CD21 ,CD 56) عند المقارنة مع النسبة المئوية لتلك الخلايا لدى مجموعة الغير متعرضين (السيطرة) بينما لم تظهر الخلايا التي تحمل المعلم المناعي (CD8) اختلافا معنوياً واضحاً عند المقارنة مع النسبة المئوية لتلك الخلايا لدى مجموعة مجموعة الغير متعرضين (السيطرة) كما اضهرت النتائج وجود انخفاضا معنويا واضحاعند المقارنة بين عن النسبة المئوية لـ CD4/CD8 لمجاميع المتعرضين عند المقارنة مع تلك النسبة لدى مجموعة الاصحاء

الاستنتاج: اشارت النتائج ان للمجال الكهرومغناطيسي تأثيرا واضحا في تثبيط الجهاز المناعي ,وذلك من خلال تأثيره على عمليات التكاثر والتمايز للخلايا المناعيه المختلفة كما ان للمجال الكهرومغناطيسي تاثيرا واضحا على المحركات الخلوية بين الخلايا المناعية حيث يعمل المجال الهرومغناطيسي على تغيير الاشارات بين المحركات الخلوية وتحويرها مما يثر سلبا على عمل النظام المناعي.

مفتاح الكلمات:المجال الكهرومغناطيسي ،الخلايا اللمفاوية ، التنميط المظهري

فرع هندسة الكيمياء الاحيائية [كلية هندسة الخوارزمي _ جامعة بغداد]

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دراسة كيمانسجية لخلايا بين العصبونات في الحبل الشوكي في اللوائن

علي عبد الستار الطائي

الخلاصة:

خلفية الدراسة: يشكل التوزيع الانظيمي للعصبونات ركن أساسي لدراسة العلوم العصبية . وقد تمت في هذه الدراسة إظهار فعالية هيدرولازات الكاربوكسيل بإستعمال الاقتران الأني بالأزو وذلك بواسطة بيوترات النافثول في خلايا بين العصبونات للحبل الشوكي للأرنب.

هدف الدراسة: تمتلك خلايا مابين العصبونات والتي تسمى خلايا رينشو فعالية التثبيط الذاتي لخلايا ألفا المحركة في الحبل الشوكي بالاضافة الى ذلك فإن هذه الخلايا تستلم إيعازات عصبية من المراكز العليا للجهاز العصبي والتي تؤثر على الافعال الانعكاسية لخلايا ألفا المحركة أما بالتسارع أو التثبيط و إن هذا الفعل قد يؤثر على أداء هذه الخلايا لما تقوم به من وظيفة حركية.

طريقة العمل: أُخذَت عينات من المادة السنجابية للحبل الشوكي من عشرة أرانب من المنطقة الظهرية- العجزية وقدعومات هذه العينات بواسطة بيوترات النافثول قبل المعاملة الاصولية لغرض الفحص بواسطة المجهر الاليكتروني النافذ.

النتائج: أظهرت النتائج فعالية واظحة و متباينة في مطرق الخلية ,المقتدرات,الشبكة الهلوية الخشنة كذلك في الهلوي المحيط بانواة للخلايا بين العصبونات والتي تم فحصها بواسطة المجهر الاليكتروني.

الأستنتاج: إن إستعمال مصطلح الخلايا المولدة ذات الطراز في الاونة الاخيرة أشارت الى خلايا مابين العصبونات في المادة السنجابية للنخاع الشوكي . ان لهذه الخلايا دور أساسي في السيطرة على حركة العضلات سواء كانت ارادية أو انعكاسية من خلال التاثير على خلايا ألفا و بيتا المحركة أو سيطرات المراكز العليا في الدماغ. أظهرت النتائج تباين واظح لفعالية هيدرولازات الكاربوكسيل باستعمال بيوترات النافثول في هذه الخلايا وبالامكان تصنيفها الى 1 و2 حسب الفعالية ومالها من دور جزيئ و جيني في تكون النوقل العصبية.

مفتاح الكلمات: خلايا مابين العصبونات ,منظمات الانماط المركزية , بيوترات النافثول ,الحبل الشوكي.

فرع التشريح [كلية الطب-جامعة النهرين]

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دراسة العلاقة بين الذيفان العصبى المستخلص من خلايا الحمضات و الربو القصبى

شهاب أحمد لافي[1] ,نضال عبد المهيمن[2] ,عامر النجار[3]

الخلاصة:

خلفية الدراسة: الذيفان العصبى المستخلص من خلايا الحمضات يعتمد فى دراسة نشاط و فعالية هذه الخلايا و كذلك فى متابعة الفعاليه الالتهابيه فى مرضى الربو القصبى.

هدف الدراسة: دراسة العلاقة بين الذيفان العصبى المستخلص من خلايا الحمضات و الربواالقصبي.

المواد و طرائق العمل: استخلص الذيفان العصبى (EDN) من خلايا الحمضات لمرضى مصابين بابيضاض الدم الحمضى (eosinophilic leukemia) .اجريت دراسة الفعاليه الحيويه لهذا المستخلص على الارانب. كما اجرى اختبار الاليزا المباشر للتاكد من وجود اجسام مضاده له فى ادرار مرضى مصابين بالربو القصبى.

النتائج: بينت النتائج ظهور اعراض ما يسمى بظاهرة كوردون على احد ارنبى التجربه خلال اليوم الثانى للحقن و انتهت بالشلل الكلى للاطراف بنهاية اليوم الخامس.اما اختبار الأليزا فقد وجد ان تركيز الذيفان اعلى فى ادرار المصابين مما هو فى مجموعة السيطره و كانت الفروقات معنويه. ولم يلاحظ فرق معنوي بين تركيزه فى ادرار مرضى الربو المصحوب بخمج بكتري ونظيره غير المصحوب بخمج .

الاستنتاجات: يمكن الاعتماد على دراسة وجود الذيفان فى الادرار فى التحرى ومتابعة مرضى الربو القصبى.

مفتاح الكلمات: الربو القصبى, الذيفان العصبىلخلايا الحمضات, اختبار الأليزا.

[1] فرع الأحياء المجهرية [كلية طب _ جامعة الأنبار]

[2] فرع الأحياء المجهرية [كلية طب _ جامعة النهرين]

[3] فرع الأحياء المجهرية [كلية طب _ الجامعة المستنصرية]

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دراسة التوصيل العصبي للعراقيين الأصحاء: بيانات طبيعية

فرقد بدر حمدان

الخلاصة:

خلفية الدراسة: دراسة التوصيل العصبي كجزء من فحوص الفسلجة العصبية الطرفية، هي امتداد للتأريخ و الفحص السريري. و هي مفيدة جدا في كل من تحديد مكان الأصابة و تحليل الحالة المرضية.

هدف الدراسة: إنشاء بيانات كهروفسلجية طبيعية للعصب القحفي السابع و اعصاب الأطراف العليا و السفلى التي يشيع اختبارها لدى أصحاء عراقيين و مقارنتها مع البيانات المنشورة في الأدبيات.

طريقة العمل: أجريت دراسة توصيل الأعصاب للأطراف العليا و السفلى في 11437 شخصا سويا من الذين تتراوح أعمارهم بين شهرين و 89 سنة.

النتائج: تم تحليل بيانات منفصلة لمختلف الفئات العمرية. في الفئة العمرية أقل من 10 سنوات، لوحظ زيادة سرعات التوصيل للأعصاب الحسية و الحركية تدريجيا مع التقدم في السن. في وقت لاحق، و في مرحلة البلوغ، انخفضت سرعة توصيل جميع الأعصاب مع التقدم في السن، و هذا واضح في كل من الطرفين العلويين و السفليين. تمت المقارنة بين نتائج العراقيين و أخرى من جميع أنحاء العالم.

الاستنتاجات: تم إنشاء معيار طبيعي لتوصيل العصب القحفي السابع و الأعصاب الطرفية العليا و السفلى في مختبر تخطيط الأعصاب في العراق. على العموم أن اختبار الأعصاب الحسية و الحركية كان مشجعا للغاية مقارنة مع البيانات الموجودة العالمية.

مفتاح الكلمات: الأطراف العليا و السفلى، الأعصاب، سرعة التوصيل، العراقيين.

فرع الفسلجة [كلية الطب _ جامعة النهرين]

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جراحة الجيوب الناظورية بالمقارنة مع الطرق التقليدية في علاج الزوائد اللحمية الأنفية والجيوب الغربالية مع الأختلالات الأنفية المصاحبة

سيوا أسعد عبد الكريم

الخلاصة:

خلفية الدراسة: هذه دراسة سريريه مقارنة أجريت في قسم الأنف والأذن والحنجرة-مستشفى السليمانية التعليمي من الأول من آب 2006 لغاية الأول من تشرين الثاني 2007 .

هدف الدراسة: أجريت هذه الدراسة لمقارنة تأثير ونتائج جراحة الجيوب الناظورية بالمقارنة مع الطرق النقليدية في علاج المرضى المصابين بالزوائد اللحمية التي تعتبر من اكثر الأنتفاخات الأنفية شيوعا.

طريقة العمل: شملت عينة الدراسة خمسون مريضا تتراوح اعمارهم بين 12 الى 75 سنة مصابون بالزوائد اللحمية الأنفية ثلاثون منهم عولجوا بالطرق الجراحية التقليدية وعشرون منهم عولجوا بجراحة الجيوب الناظورية.تم متابعة المرضى بعد اجراء العمليات الجراحية باستخدام معايير الأعراض المرضية والفحوصات الناظورية0

النتائج: أظهرت النتائج إن جراحة الجيوب الناظورية ادت الى تحسن افضل في الأعراض وعلاج الأختلالات الأنفية المصاحبة مع انخفاض نسبة المظاعفات ونسبة تكرار الحالة المرضية.

ومن جهة اخرى تتطلب جراحة الجيوب الناظورية خبرة عالية ووقت اكثر لأجرائها من الطرق التقليدية.

الاستنتاج: استنتجنا من الدراسة ان جراحة الجيوب الناظورية افضل من الطرق الجراحية التقليدية في علاج الزوائد اللحمية الأنفية لأنها تعطي مجال رؤيا اقرب مع اضاءة افضل مع الوصول الى موضع الخلل بشكل افظل وان الأختلالات الغير ظاهرة ممكن الوصول اليها وعلاجها وان المضاعفات وتكرار حدوث الحالة المرضية نسبتها اقل.

مفتاح الكلمات: الزوائد اللحمية الأنفية,استئصال الزوائد اللحمية التقليدي,جراحة الجيوب الناظورية.

فرع الأنف والأذن والحنجرة-مستشفى السليمانية التعليمي

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فرط ضغط الدم الرئوي عند المصابين بالعجز الكلوي المزمن

جواد كاظم مناتي

الخلاصة:

خلفية الدراسة: فرط ضغط الدم الرئوي يوجد عند المصابين بالانسداد الرئوي المزمن وأمراض التليف الرئوي وكذلك أمراض جدار القفص الصدري. وتبين أن المصابين بالعجز الكلوي يعانون من فرط ضغط الدم الرئوي والذي يؤدي إلى عجز القلب ولذلك تظهر أعراض ضيق التنفس.

هدف الدراسة: تهدف هذه لدراسة لتقييم معدلات انتشار فرط ضغط الدم الرئوي عند المصابين بالعجز الكلوي قبل البدء بالديال الدموي وأثناء الديال الدموي مع بحث عن العوامل التي تؤدي إلى زيادة فرط الدم الرئوي .

طريقة العمل: تضمنت الدراسة مائة مريض مصابين بالعجز الكلوي. خمسون تحت العلاج التحفظي بدون غسل كلوي وخمسون يعالجون بالديال الدموي من عام 2008الى 2009 .تم تقييم معدلات انتشار فرط ضغط الدم الرئوي بشكل استباقي مستخدمين تصوير القلب بالأمواج فوق الصوتية الدوبلر وإجراء فحوصات الدم مثلا نسبة الهيموكلوبين , البروتين, اليوريا, كالسيوم

النتائج: تم تشخيص ارتفاع ضغط الدم الرئوي عند المصابين بعجز الكلى المزمن(أعلى من 35 ملم/زئبقي)عند33(33%) مريضا .21 مريض يعالجون بالديال الدموي و 12 مريضا يعالجون بالعلاج التحفظي(بدون ديلزة)

الاستنتاجات: تشير هذه الدراسة لارتفاع معدلات انتشار فرط الضغط الرئوي عند مرضى العجز الكلوي المزمن ومن الضروري الكشف المبكر لهذه المضاعفات بهدف تفادي العواقب المترتبة على ذلك.

مفتاح الكلمات: فرط ضغط الم الرئوي، الديال الدموي ، قبل الديال، دوبلر

فرع الباطنية_شعبة الديلزة[كلية الطب_جامعة النهرين/ مستشفى الكاظمية التعليمي]

المجلة العراقية للعلوم الطبية 2009 م المجلد 7 العدد2 ص104-108

دراسة علم الشكل(مورفومتريه) للتغيرات في صبغة نترات الفضة في الخلايا القاطنة في العضلات الهيكلية مع التقدم بالعمر

مي فاضل ماجد الحبيب , هدى رشيد كريم

الخلاصة:

خلفية الدراسة: العضلات الهيكلية من انسجه الجسم التي تتأثر بتقدم العمر.إن تأثيرات تقدم العمر في العضلات الهيكلية لم تتم دراستها بالربط مع صبغة نترات الفضة.

هدف الدراسة: هذه الدراسة تهدف لتوضيح تأثيرات تقدم عمر العضلات الهيكلية في صبغه نترات الفضة باستخدام علم الشكل و طريقة العد.

طريقة العمل: تمت دراسة نماذج من العضلة الباسطة للأصابع الطويلة لأربعين جرذ من الذكور, وبأعمار تتراوح بين 27 يوما إلى 18 شهرا. حضرت قوالب شمعيه من النماذج وقطعت إلى شرائح بسمك (5-6)مايكرون. استخدمت هذه الشرائح لصبغه نترات الفضة . تم تحليل النتائج عن طريق احتساب عدد النويات للخلية المصطبغة بصبغه نترات الفضة و باستخدام برنامج (التصوير ألمختبري الشامل الثاني) والذي فيه يتم ربط المجهر بجهاز كومبيوتر تم تحليل الصور التي ترى من خلال المجهر واحتساب منطقه النواة, ومحيط النواة وكرويتها.

النتائج: المجموعة العمرية لحديثي الولادة أظهرت الانويه ألفه عاليه اتجاه الصبغة, وأعلى عدد من النويات لمصبغه نترات الفضة

المجموعة العمرية البالغة قلت الالفه اتجاه الصبغة, وظهرت الانويه كنقاط صغيره.وتم تمييز نسبه عاليه من الانويه في المقطع الواحد.

المجموعة العمرية الكبيرة قلت كثافة الصبغة بصوره ملحوظة, وتحتوي النواة على صبغه نترات الفضة منفردة ومنعزلة وكرويه.

الاستنتاجات: هنالك اختلافات مهمة في صبغه نترات الفضة للخلايا القاطنة في العضلات الهيكلية (الانويه العضلية والخلايا التابعة) مع تقدم العمر تظهر من خلال وجود نشاطات انقساميه وفعالية المتغيرات(منطقه النواة ومحيطها وكرويتها).

مفتاح الكلمات: العضلات الهيكلية, صبغه نترات الفضة.

فرع التشريح _ شعبة الأنسجة والأجنة [كلية الطب_جامعة النهرين]

المجلة العراقية للعلوم الطبية 2009 م المجلد 7 العدد2 ص109-115

توازن العناصر الضئيلة لدى الحوامل المصابات بارتفاع ضغط الدم (قبل الشنج)

فيصل غازي الربيعي

الخلاصة:

خلفية الدراسة: ضغط الدم العالي لدى الحوامل (قبل الشنج) هو نوع من ارتفاع ضغط الدم يظهر أثناء الحمل, وهو من المضاعفات الشائعة المؤدية إلى نسبة وفيات ومضاضة عاليتين؛ ومع ذلك سبب هذا الارتفاع غير معلوم. المعطيات حول أتزان معادن الدم والعناصر الضئيلة وخاصة

الايونـات الموجبـة أثنـاء الحمـل تعتبـر متناقضـة؛ إضـافة لـذلك, فـأن حالـة أيونـات الكالسـيوم والمغنيسيوم الحرة خلال الحمل ومضاعفاته (ارتفاع ضغط الدم المصاحب للحمل "قبل الشنج") لم توصف بشكل كامل.

هدف الدراسة: بيان نمط المعادن (الكالسيوم و المغنيسيوم) و العناصر الضئيلة (الخارصين و النحاس) في حالة ارتفاع ضغط الدم المصاحب للحمل (قبل الشنج) وعلاقته مع الحمل الطبيعي.

طريقة العمل: هذه الدراسة هي دراسة مقطعية تشمل قياس المعادن (الكالسيوم, المغنيسيوم) لدى 60 حاملا" مصابة بارتفاع ضغط الدم المصاحب للحمل (مجموعة الاختبار) وتم تصنيفهم إلى مجموعتين حسب عمر الحمل:

حوامل مصابات بارتفاع ضغط الدم المصاحب للحمل (قبل الشنج) خلال الفصل الثاني من الحمل (العدد30 مريضة).

حوامل مصابات بارتفاع ضغط الدم المصاحب للحمل (قبل الشنج) خلال الفصل الثالث من الحمل (العدد30 مريضة).

تمت مقارنة النتائج مع نتائج 60 حاملة" سليمة" ظاهريا"(مجموعة السيطرة)، وتم تقسيم مجموعة السيطرة اعتمادا" على عمر الحمل إلى مجموعتين:

حوامل صحيحات ظاهريا" خلال الفصل الثاني من الحمل (العدد30 مريضة).

حوامل صحيحات ظاهريا" خلال الفصل الثالث من الحمل (العدد30 مريضة).

النتائج: أظهرت تغيرا" في شوارد المصل والعناصر الضئيلة لدى الحوامل ذوات ضغط الدم العالي المصاحب للحمل (قبل الشنج) يشمل انخفاضا معنويا لمستوى المغنيسيوم والكالسيوم المصحح للألبومين في الدم ، وانخفاضا في الخارصين المرتبط بارتفاع معنوي لمستوى النحاس في مصل الحوامل المريضات لدى مقارنتهن بمجاميع السيطرة المناظرة.

الأستنتاج: إن الحوامل ذوات الضغط العالي المصاحب للحمل (قبل الشنج) يعانين من اختلال حالة توازن معادن الدم والعناصر الضئيلة عند مقارنتهن مع الحوامل الصحيحات المناظرات للمريضات في العمر ومدة الحمل.

جميع الحوامل المصابات بارتفاع ضغط الدم المصاحب للحمل (قبل الشنج) كانت لديهم بعض العوامل التي تثبط توسع الاوعية الدموية وتزيد انقباضها وتحدث إجهادا تأكسدياً ومما يدل على هذا وجود انخفاض معنوي في مستوى المغنيسيوم الحر(الذي يعتبر ضروريا" للحفاظ على انبساط الوعاء الدموي) والكالسيوم الحر(الذي يعتبر ضروريا" لتكوين اوكسيد النتريك المشتق من بطانة الاوعية الدموية) وأرتفاع النحاس (الذي يولد انواعا من الاوكسجين الشديدة التفاعل) مع انخفاض الخارصين(الذي يعتبر ضروريا كمادة مقاومة للتاكسد).

مفتاح الكلمات: قبل الشنج، الكالسيوم، المغنيسيوم، النحاس، الخارصين.

فرع الكيمياء و الكيمياء الحياتية [كلية الطب _ جامعة النهرين]

المجلة العراقية للعلوم الطبية 2009 م المجلد 7 العدد2 ص116-123

ورم الجوف القحفي التصادفي "تقرير حالة "

معتز عبد المجيد عبد العزيز القزاز

الخلاصة

أجريت دراسة هذه الحالة على جثة امرأة بلغت من العمر خمسين عاما , تمت احلتها من قبل الجهات الحقيقية إلى معهد الطب العدلي ببغداد كحالة وفاة بحادث طريق لغرض تشريحها كان التاريخ المرضي لحالتها الصحية قبل الوفاة سالبا اعتمادا على معلومات تم الحصول عليها من

ذويها أظهر التشريح الطبي العدلي الاصولي وجود ورم في داخل الجوف القحفي عند قاعدة الدماغ وبعد إجراء الفحص ألمجهري النسيجي للورم تبين انه ورم سحائي.

مفتاح الكلمات: ورم الجوف القحفي , ورم سحائي , تشريح الجثة , ورم الدماغ.

فرع الطب العدلي [كلية الطب _ جامعة النهرين]

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